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STUDIES ON CEREAL RUSTS

I. *PUCCINIA PENNISETI* ZIMM. AND ITS ALTERNATE HOST

By T. S. Ramakrishnan and C. K. Soumini

(Accepted for publication October 8, 1948)

PUCCINIA PENNISETI was first recorded by Zimmerman from Amani, East Africa, in 1904 on the leaves and culms of *Pennisetum typhoides* Stapf & Hubb. Barclay used the name *Puccinia penniseti* Barclay, in 1890, which according to Butler & Bisby (1931) was without formal description and which he had confused with *Puccinia purpurea*. The same rust was observed on this host by Butler (1918) from several parts of India. Outside India it is recorded from Africa, where it is known to infect other species of the genus *Pennisetum*.

Fig. 1. Section through uredium; urediospore and its germination (x 250)

The rust is common in many districts of the Madras province on *Pennisetum typhoides* (cumbu or bajra). The incidence of the rust is evident in the crops raised in different seasons of the year. Usually the uredia begin to develop at the time the crop is about to flower though in some years they have been observed much earlier. The sori first appear in the upper half of the leaves nearer the tip. Soon, however, the intensity of infection increases and they are found throughout the leaves and in severe cases, cover the entire surface. Uredia are deep yellowish brown and present on both sides of the leaf, being more common on the upper surface. Telia develop much later than the uredia. They are smaller, isolated or in groups and long remain covered by the epidermis. Very often the tissues of the leaf around the sori quickly dry up with the result that the latter are seen in the middle of straw coloured lesions. The whole leaf may wither completely; when the incidence of the rust is heavy, the field presents a scorched appearance.

UREDIA

The uredia are formed subepidermally. At maturity they rupture the epidermis which forms flakes round the sori and the spores are then exposed. The

urediospores are borne on hyaline pedicels which extend up to 60μ in length. Butler (1918) states that the pedicels are shorter than the spores. However, in the specimens examined by us, they are much longer. The urediospores are oval,

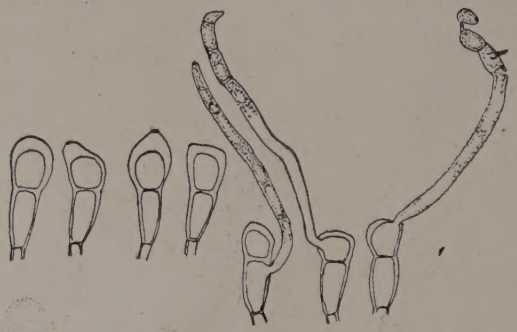


FIG. 2. Teliospores and their germination (x 250)

pyriform or elliptic with four equatorial germ pores and measure 35 x 25μ (25-42 x 21–30μ). The spores are yellowish brown with a coloured wall and sparsely echinulate, the echinulations being abundant near the apex. Butler (1918) has stated that paraphyses are absent. But in a number of sori examined by us groups of paraphyses have been seen mainly along the margin. These are cylindric or slightly inflated at the apex, flexuous, incurved and stouter and longer than the pedicels. Sometimes they are faintly tinted brown. These paraphyses are not clearly visible in scrapings but can be readily recognised in sections. The number of paraphyses varies in different sori.

TELIA

The telia are amphigenous, often in groups and black. They are formed subepidermally and remain covered by the epidermis for a long time without bursting. In many cases the sori are divided into two or more compartments by walls of sterile tissue. These partitions are cellular and do not resemble paraphyses. The teliospores are dark brown in colour, elongated, cylindric to club-shaped, broad in the upper portion and tapering towards the stalk, with the lower cell longer than the upper one. The apex is flattened or rounded, sometimes blunt, thickened up to 10μ and often darker in colour than the rest of the walls. The spores measures 49 x 21μ (33-59 x 13-30μ). The pedicel is very short and measures on an average 9μ (4-19μ). It is coloured, though Butler (1918) states that it is colourless.

SPORE GERMINATION

Urediospores germinate readily producing one or more germ tubes. The germ tubes are stout, flexuous, sometimes branching and often enlarged at the tip. The viability of the spore is, however, limited. Soon after collection, germination is very high, with 90-100% viable spores. But the spores on air-dried specimens kept at the laboratory temperature (28-30°C) in thin paper envelopes gradually lose their viability and at the end of 30 days no viable spore is found. There is quicker loss of viability when the specimens are stored under conditions of high or very low humidity (75 to 100 per cent or below 5 per cent relative humidity). Spores from material

stored at about 40 to 50 per cent relative humidity germinate up to 20 days of storage but not later, while in the former instances the viability is lost in 10 to 15 days.

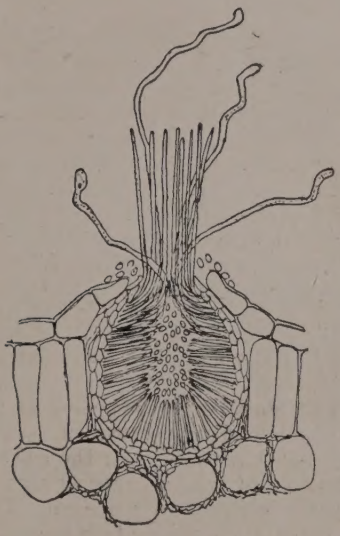


FIG. 3. Section through pycnium (x 250)

Fresh teliospores exhibit 5 to 10 per cent germination when kept in drops of sugar solution for 72 to 100 hours. In most cases the promycelium develops from one cell only either the upper or the lower. It is long and septa are developed nearer the apex. One sterigma is formed from the upper end of each cell and on each of these an elliptical or round basidiospore is borne.

INFECTION EXPERIMENTS ON BAJRA

Inoculations were carried out with the urediospores on the leaves of bajra. Healthy seedlings 3 to 4 weeks old and grown from disinfected seed were utilised. Urediospores were obtained from fresh collections and transferred by means of a brush or scalpel to the surface of leaves. The inoculated plants were kept in an isolated area and covered by bell jars for 72 hours after inoculation. Sori were formed in 8 to 10 days. The three strains of bajra viz. Co. 1, Co. 2 and Co. 3, strains evolved at the Millets Breeding Station, Coimbatore, for economic characters were almost equally affected by the rust as revealed by field observations and artificial inoculations. Leaves of *Paspalum scrobiculatum* L. and *Digitaria marginata* Link were also inoculated with the urediospores but no signs of infection developed on either of them at any time. In all experiments the control plants remained free from rust.

ALTERNATE HOST

Butler (1918) states that the æcial stage of this rust is unknown and that probably the rust has dispensed with this stage. Several unrelated æcia occurring on *Ipomœa* spp. *Solanum* spp., *Diospyros melanoxylon* Roxb., etc. are prevalent in the province. The presence of germinating teliospores suggested that the basidiospores

may be inoculated on the leaves of some of these plants usually found round the bajra fields to see if there is any relation between *Puccinia penniseti* and the æcia found on them.

At the outset, *Oxalis* sp. *Blepharis bœrhaaviæfolia* Pers., *Ipomœa reptans* Poir. and *Solanum melongena* L. were tried. Inoculations were made in the manner described by Arthur (1929). Healthy leaves were removed, washed in several changes of distilled water and placed in sterilised petri dishes containing small quantities of sterilised water. Basidiospores and germinating teliospores were transferred to the upper surface of the leaves. The water in the dish was replaced by one percent sugar solution on the third day and further changed daily. By this means it was possible to keep the leaves in a fresh condition for 8 to 10 days. The initial experiments with *Solanum melongena* alone were successful. On the eighth day small swellings developed on the lower surface of the leaf of *Solanum melongena* and on the tenth day pycnia and æcia were observed.

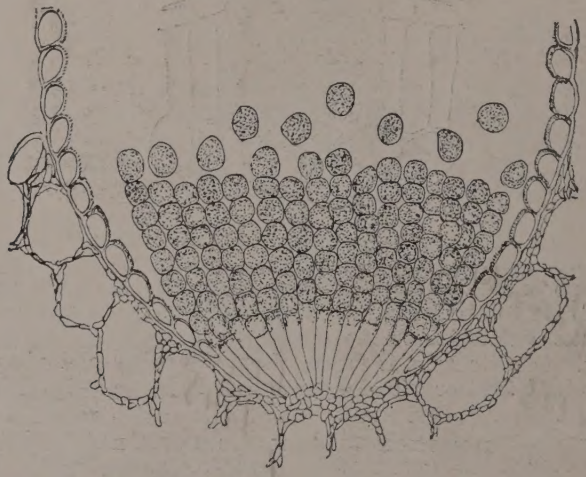
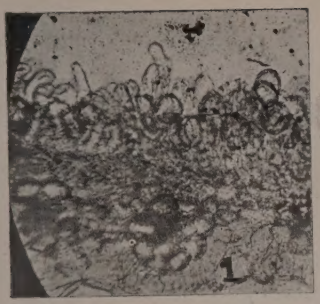
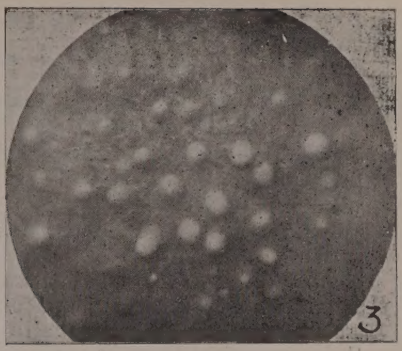


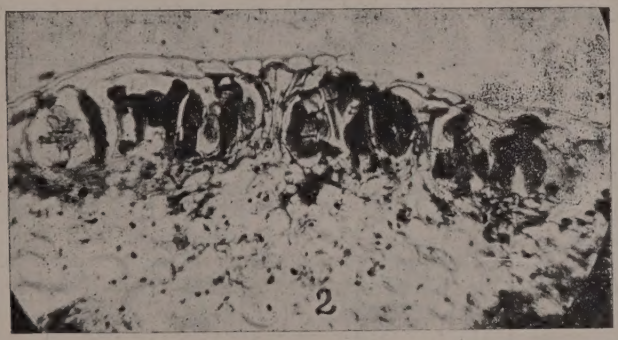
FIG. 4. Section through æcium (x 250)

Infection experiments with the basidiospores were repeated several times on the leaves of seedlings and older plants of *Solanum melongena* growing in pots. In all of them numerous pycnia and æcia developed. The infected portion of the leaf was light green, thickened and usually concave on the upper surface with a corresponding convex bulge on the lower surface. The spots gradually increased in size to a maximum of 2 cms. in diameter. Sometimes the petioles were also affected. Pycnia invariably developed on the upper surface and appeared as orange yellow, minute dots with glistening drops of nectar. The pycnium was subepidermal, 110 x 140μ, paraphysate and deep orange in colour. The paraphyses were straight and coloured. Mixed with these were long, hyaline, flexuous hyphæ. Pycniospores were oblong or elliptic, hyaline and measured 4-8 x 2-3μ.

Æcia were produced on the lower surface but very rarely one or two æcia could be seen on the upper surface also. On the petioles they were formed on all sides. These were arranged in irregular, concentric expanding circles. Each æcium was salmon orange in colour, columnar and projecting beyond the surface to a height of







1. Uredium showing paraphyses (x 150)
2. Secting of telium with cellular partitions (x 250)
3. Young aecia on *Solanum melongena* (enlarged)
4. Groups of aecia (nat. size)
5. Aecia on petioee (nat. size)

1mm. There was a distinct peridium but this did not open well or become lacerated as is common in other æcia ; irregular splits occurred at the tip. More often the whole æcium dropped off as a short rod. The peridium was made up of hyaline, polygonal cells 23 x 20μ (16-30 x 14-25μ), prominently verrucose on the inner surfaces. The æciospores were catenulate, formed on a prominent fertile layer of elongated cells. The spores were globose to angular, yellowish orange, thin walled smooth, and measured 21 x 18μ (16-25 x 12-21μ). The æciospores were inoculated on the leaves of young bajra plants, three weeks old. Urediosori developed in the course of 8 to 10 days. The control plants were quite healthy and free from rust. It was thus possible to follow the completion of the life cycle of the rust. These experiments have definitely proved that this rust is heterœcious and that the brinjal plant, *Solanum melongena*, functions as an alternate host. This plant is widely cultivated in the *bajra* growing tracts and there is little doubt that it serves as an alternate host in nature.

DISCUSSION

Puccinia paspalicola (Pat. & Gaill) Arth. develops its æcial stage on *Solanum melongena* (Arthur 1934, Mundkur 1938, Saccardo 1891, Ocfemia 1935). But the æcia are longer and the spore size is different from those of the rust under study. Furthermore infection experiments with æciospores of the present rust and with the urediospores of *Puccinia penniseti* on the leaves of *Digitaria marginata* and *Paspalum scrobiculatum* gave negative results thereby establishing the differences between the rusts. *Æcidium solani* Mont. (Butler & Bisby 1931, Saccardo 1888) has been observed on the leaves of *Solanum* sp. and is different from the æcial stage obtained on *Solanum melongena*. It has to be concluded that *Solanum melongena* serves as an alternate host for more than one species of rust.

Puccinia penniseti apparently exhibits wide variation in its characters, as seen from a comparison with the earlier descriptions of the rust given by Butler (1918). The local collections were compared with specimens of *Puccinia penniseti* obtained from the Commonwealth Mycological Institute, Kew, through the kind courtesy of Dr G. R. Bisby. The hosts were *Pennisetum leonis* Stapf & Hubb. and *Pennisetum typhoides* and had been collected in Africa. The rusts on these were identical with the one under study. An emended description of the rust including the pycnial and æcial stages on the alternate host is given below.

Puccinia penniseti. Rust spot hypertrophied, thickened, greenish yellow. Pycnia epiphyllous, subepidermal, ovate to globose, paraphysate, 110 x 140μ, orange yellow ; pycniospores, hyaline, oval to oblong 4–8 x 2-5μ.

Æcia hypophyllous, rarely epiphyllous, sometimes on the petiole, arranged in irregular concentric circles, columnar, up to 1mm. long, salmon orange ; peridial cells hyaline, polygonal, verrucose, 23 x 20μ (16–30 x 14–25μ) ; *æciospores* catenulate, globose to angular, yellowish orange, smooth, 21 x 18μ (16–25 x 12–21μ).

Uredia amphigenous, mostly epiphyllous, brown, subepidermal, erumpent, paraphysate, paraphyses on the margin of the sorus ; *urediospores* pedicellate, pedicel hyaline, of varying lengths up to 60μ, oval, elliptic or pyriform, yellowish brown, echinulate, with four equatorial germ pores, 35 x 25μ (25–42 x 21–30μ).

Telia amphigenous, in groups, black, subepidermal, long covered by epidermis, often divided into compartments ; teliospores dark brown, pedicellate, with short

con-colorous pedicel, elongated, cylindric to club-shaped, 49 x 21μ (33–60 x 13–30μ) apex various, thickened up to 10μ, constricted at the septum, smooth.

Uredia and telia on leaves and stem of *Pennisetum typhoides, Pennisetum leonis,* and other species of *Pennisetum.*

Pycnia and æcia on leaves and peticles of *Solanum melongena.*

We gratefully express our thanks to Dr G. R. Bisby for kindly supplying specimens and giving helpful suggestions.

SUMMARY

Fresh collections of *Puccinia penniseti* on *Pennisetum typhoides* were examined and found to have long pedicels on the urediospores, paraphyses in uredia and compartments in telia, which features have not been recorded before. *Solanum melongena* was found to serve as an alternate host on which pycnia and æcia were produced by infection with basidiospores and the æciopores passed on to *bajra.* This is the first reord of these stages for this rust. A complete and emended description is now provided.

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BREMIA SP. ON ARTHRAXON LANCIFOLIUS HOCH IN INDIA

By M. K. Patel

(Accepted for publication October 14, 1948)

SINCE the establishment of genus *Bremia* and the species *Bremia lactucae* by Regel in 1843, several species belonging to the family Compositae have been found to be attacked by this fungus. Chief among the genera whose species are attacked, are *Carthamus*, *Centaurea*, *Cichorium*, *Conyza*, *Crepis*, *Cynara*, *Gaillardia*, *Lactuca*, *Saussurea*, *Senecio*, *Sonchus*, *Taraxacum*, etc. The genus *Bremia* has been studied by Büren (1915, 1922) in Switzerland who, following the lead of Ed. Fischer and his school, has erected several "Kleine" species. But they are all on species of Compositae. Outside the family, the species of this genus have been reported on only very few families. Naoumoff (1914) found a *Bremia* on *Arthraxon ciliaris* in Austria, Oriental Russia and China. The conidia of the fungus were, according to Naoumoff (1914) 12μ in diameter and he named it *Bremia graminicola* Naoumoff. Togashi (1926) recorded it for Japan on the same host but he stated that the conidia are papillate and 11 to 15μ in diameter. Both Naoumoff and Togashi state however that the conidia are round.

While collecting fungi on the hills near Mahableshwar, a species of *Bremia* was found on *Arthraxon lancifolius* in October 1942. A cursory examination of the fungus indicated that its conidia are very large. A careful study of the morphology of the fungus was therefore made in order to precisely determine it. Twenty one conidiophores and a hundred conidia were measured using a filar micrometer under artificial light. The conidiophores with their characteristic disc-like structures bearing sterigmata, ranged from 332.5 to 840μ in length with an average of 493.5μ. The conidial measurements are given in Table I.

Table I

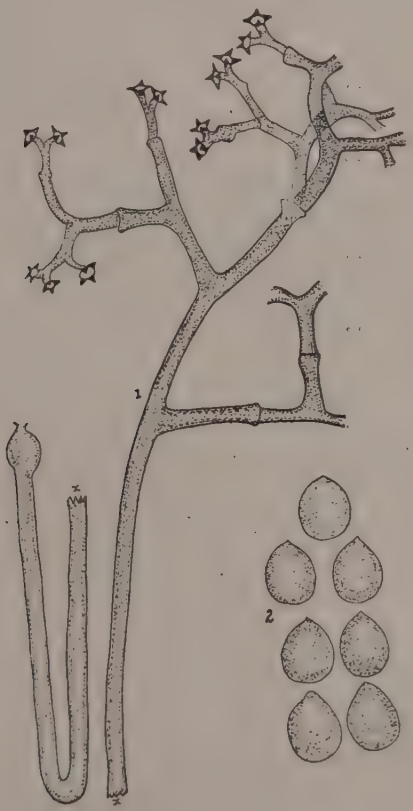
Measurement of conidia of Bremia *on* Arthraxon

Length		Breadth	
Classes in μ	No. in 100	Classes in μ	No. in 100
11–11.99	7	9– 9.99	32
12–12.99	14	10–10.99	30
13–13.99	14	11–11.99	33
14–14.99	23	12–12.99	5
15–15.99	21		
16–16.99	14		
17–17.99	7		
Mean	14.5μ		10.6μ

The conidia, unlike those of *Bremia graminicola* which are round, are distinctly ovate and range from 11 to 18μ in length and 9 to 13μ in breadth with an average of 14.5 x 10.5μ as compared to 12μ diameter stated by Naoumoff and 11–15μ by Togashi for *Bremia graminicola*. Apparently the Mahableshwar fungus is not the same as the one occuring in China, Japan, Russia or Austria and is proposed as a new variety of *Bremia graminicola*.

Bremia graminicola Naoumoff var. **indica** var. nov.

Spots large or small, woolly, whitish to yellowish brown, occupying a portion of the entire leaf surface; conidiophores hypophyllous, sometimes epiphyllous, caespitose, floccose, hyaline with a bulbous base; branches arising alternately 6-7 times from the main axis; lowest branch starting from above half the length of the conidiophore, then branching dichotomously; end in disc-like structures with 4 sterigmata; rarely the main conidiophore, more often branches, thickens and forms septa; measuring 332.5—840 x 8—10.5μ. Conidia ovate, hyaline, with distinct papilla, measuring 11—18 x 9.1μ. Oospores not seen.



FIGS. 1 & 2

On living leaves of *Arthraxon lancifolius* Hoch from Mahableshwar, Bombay Province, October 1942, leg. M. K. Patel. Type deposited in the Herbaria of the College of Agriculture, Poona, of the Indian Agricultural Research Institute, New Delhi, and of the Commonwealth Mycological Institute, Kew, England.

Bremia graminicola var. **indica** var. nov.

Differt a specie habens ovata conidia 11—18μ longitudine et 9—13μ latitudine. Collecta a M. K. Patel mense Octobri 1942 in Mahableshwar, Bombay.

Differs from the species in having ovate conidia measuring 11—18μ in length and 9—13μ in breadth.

The author wishes to express his appreciation to Dr B. N. Uppal, Director of Agriculture, B. P., Poona and Dr B. B. Mundkur for the help given during the progress of this work and in preparing the manuscript of this paper; and to Rev. Fr. M. Singarayer, Coimbatore, for the latin diagnosis of the new variety.

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Togashi K. (1926) ..Notes on some parasitic fungi of Japan. *Bull. Imp. Coll. Agric. Forestry* (Morioka) **9**: 17–29.

Explanation of figures

Figs. 1 *Bremia graminicola* var. *indica* nov. var. Conidiophore with bulbous swelling, branching indefinitely, dichotomous; terminal branchlet with shallow saucer-shaped disc bordered by 4 sterigmata, each bearing a conidium. x 600. Fig 2. Conidia hyaline, ovate and lacking characteristic apical papilla x 1600.

A THIRD CONTRIBUTION TOWARDS A KNOWLEDGE OF INDIAN USTILAGINALES

(Fragments : LI—LXXXVI)

By M. S. Pavgi and B. B. Mundkur

(Accepted for publication October 15, 1948)

(This is the third and last contribution in the series ; the first two were published in the *Trans. Brit. Mycol. Soc.*, **23** : 86—121, 1939 and **24** : 312—336, 1940. B. B. M.)

LI **Cintractia axicola** (Berkeley) Cornu, *Ann. Sci. Nat. Bot.* 6, **15** : 279, 1883 ; Saccardo, *Syll. Fung.* **7** : 480, 1888 : Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 43, 1931.

Syn. *Ustilago axicola* Berkeley, *Ann. Mag. Nat. Hist.* 2, **9** : 200, 1852.
Ustilago fimbristylis Thuemen, *Bull. Torrey Bot. Cl.* **6** : 95, 1876
Cintractia mundkuri Chowdhury, *Indian J. Agric. Sci.* **14** : 231, 1944.

Sori usually at base of peduncles or pedicels, rarely in spikelets, globular or elongate, 2-4mm. ; covered at first with pinkish-grey pseudo-membrane ; this later flakes away exposing the inner olive-black firmly agglutinated spore-mass ; sterile cells indefinite through gelatinization and filiform. Spores chestnut-brown (Ridgway), spherical, broadly oval, laterally compressed, 12—18.5μ in diameter with a mean of 15μ ; epispore dark, very thick, giving a minutely punctuate appearance due to granular spore contents.

On *Fimbristylis quinquangularis* Kunth at Yelwal, Mysore. Collected by E. J. Butler on September 9, 1904 ; on *Fimbristylis annua* R. and S. var. *diphylla* Kuk. at Mukkamala, Godavary (S. N. Mitra on September 12, 1907) ; on *Fimbristylis* sp. Erramacola, Wynaad (W. McRæ on September 18, 1909 ; at Nagpur (P. A. Pandit on September 21, 1908) ; on *Fimbristylis diphylla* Vahl. at Habiganj, Assam (S. Chowdhury on March, 7, 1942) ; on *Fimbristylis dichotoma* Vah. at Amritsar, Punjab (A. Hafiz Khan on October 12, 1907) ; on *Fimbristylis tenera* R. and S. at Rohtak, Punjab (S. Ahmad on November 7, 1942).

LII **Cintractia caricis** (Persoon) Magnus, *Abh. Bot. Ver. Brand.* **37** : 79, 1896 ; Clinton and Zundel, *Mycologia* **30** : 281, 1938.

Ovariicolous ; sori at first hidden by outer glumes and exposed as globoid occasionally elongate bodies, 1—2mm. in size, covered by a whitish papery pseudomembrane ; this soon flakes away disclosing the black agglutinated spore-mass with a short columella sometimes protruding out of the sorus. Spores carob-brown (Ridgway), subopaque, irregularly polyhedral, subglobose, elongate in lateral view, 16.5—24μ in diameter with a mean of 21μ ; epispore dark, very thick, verrucose, sometimes papillate.

On *Carex cardiolepsis* Nees at Sonamerg, Kashmir. Collected by R. R. and I. D. Stewart on July 23, 1921.

LIII **Cintractia fimbristylicola** sp. nov.

Ovariicolous ; sori enclosed within the glumes and later exposed, globoid, 1—2mm. in size, covered by a whitish, papery, pseudo-membrane ; membrane later flakes away disclosing the black, firmly agglutinated spore-mass. Spores chestnut-brown (Ridgway), spherical, broadly oval, laterally compressed, 7—11μ in diameter with a mean of 9μ ; epispore very thick, dark, verrucose, occasionaly having mycelial remnants.

On *Fimbristylis complanata* Link at Chatrapur, Ganjam, Orissa. Collected by E. J. Butler on August 30, 1904.

Ovariicola ; sori inclusi in glumis, tandem expositi, globoidei, magnit, 1—2mm, operti albescente papyracea membrana quæ postea flocculatim dehiscit, expositis nigris sporarum massis firmiter aglutinatis. Sporæ castaneo-brunneæ sphæricæ, late ovatæ, lateraliter compraassae, 7—11μ diam. medietate 9μ ; episporium crassissimum, obscurum, verrucosum ; sporæ nonnumquam mycelii reliquiis ornatæ.

Habitat, in inflorescentia Fimbristylis complanatæ Link ; leg. E. J. Butler, Chatrapur, Ganjam, Orissa, 30-8-1904. Typus.

LIV **Cintractia inclusa** (Brefeld) Liro, Die Ustilagineens Finnlands, **2** : 16, 1935.

Syn. *Anthrocoidea inclusa* Brefeld, *Unters. Gessamt. Mykol.* **15** : 36 and 150, 1912.

Sori in ovaries, covered by outer glumes and then exposed, globoid to elongate, 1—2.5mm. ; enveloped in a thin whitish pseudo-membrane ; membrane later wears away disclosing the semi-agglutinated, blackish-brown spore-mass with a stout columella. Spores Brusse's brown (Ridgway), subglobose, often elongate or broadly oval with blunt angles, 13—20.5μ in diameter with a mean of 16μ ; epispore thick, verrucose, giving a punctate appearance due to granular spore contents.

On *Carex stenophylla* Vahl. above Losar (Spiti). Collected by S. Ahmad on September 20, 1935.

LV **Cintractia leioderma** (Lagerheim) Cifferi, *Ann. mycol. Berl.* **29** : 72, 1931.

Syn. *Ustilago caricis* var. *leioderma* Lagerheim, *Mitt. Bad. Bot. Ver.* p. 37, 1888.

Sori infecting ovaries, covered at first by glumes and then exposed, globular to slightly elongate, 1.5—2.5mm. ; enveloped by a thin whitish pseudo-membrane ; membrane later flakes away exposing the firmly agglutinated, black spore-mass ; stout columella present, rarely protrudes out of the sorus. Spores chestnut-brown (Ridgway), spherical, globose to broadly oval, 15—20μ in diameter with a mean of 18μ ; epispore very thick, ferruginous (Ridgway), and smooth.

On *Carex incurva* Lightf. below Babeh Pass (Bashahr). Collected by S. Ahmad on October 6, 1935.

LVI **Cintractia peribebuyensis** (Spegazzini) Spegazzini, *An. Soc. Cient. Argent.* **26** : 9 (of reprint), 1888 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 44, 1931.

Syn. *Ustilago peribebuyensis* Spegazzini, *An. Soc. Cient. Argent.* **16** : 48 (of reprint), 1883 ; Saccardo, *Syll. Fung.* **7** : 458, 1888 .

Cintractia axicola var. *minor* Clinton, *J. Mycol.* **8** : 143, 1902.

Cintractia minor Jackson, *Mycologia*, **12** : 153, 1920.

Cintractia distans Mundkur, *Indian J. Agric. Sci.* **14** : 50, 1944.

Sori usually formed at base of peduncles, occasionally in spikelets, covered by glumes, elongate, slightly tapering upwards, 3—8 mm. long, covered by a pink, rather tough pseudo-membrane ; membrane later wears away exposing the olivaceous-black, agglutinated spore-mass. Spores chestnut-brown to Mar's brown (Ridgway), spherical, globose to broadly oval, sometimes angular, laterally compressed, 9—13μ in diameter with a mean of 11μ ; epispore thick, ferruginous (Ridgway), giving punctate appearance due to highly granular spore contents.

On *Cyperus rotundus* Linn. at Yellareddy, Hyderabad, Dn. collected by S. Vaheeduddin on August 21, 1941 : on *Cyperus sp.* at Pusa (C. B. Sahaya on August 24, 1931) ; at Awapur, Dist. Muzaffarpur (Jamal Bux on October 7, 1911) ; at Dhalghat, Chittagong (R. Sen on August 22, 1911) ; at Almore, Godavary (L. S. S. Mony on November 25, 1910) ; at Samalkot, Madras (F. J. F. Shaw on November 21, 1910) ; at Nagpur (P. A. Pandit on October 30, 1908) ; at Lonavala, Bombay (Prof. Saxton) ; at Bilikere, Mysore (E. J. Butler on September 19, 1903) ; on *Cyperus distans* Linn. at Dacca, Bengal. (P. Maheswari in August, 1940, type of *Cintractia distans*.)

LVII **Cintractia peribebuyensis** Spegazzini var. **major** var. nov.

Sori at the base of peduncles of inflorescence, globular to elongate or conical, 2—5mm. long, 0.5—2mm. broad, covered by a pinkish pseudo-membrane ; membrane later wears away exposing olivaceous-black, agglutinated spore-mass. Spores chestnut-brown (Ridgway), spherical, globose to broadly oval, laterally compressed, 13—18μ in diameter with a mean of 15μ ; epispore very thick, dark ferruginous (Ridgway), smooth, with highly granular contents.

On *Cyperus* sp. at Cocanada, Madras. Collected by S. Sundararaman in December, 1906.

Sori ad basim pedunculorum vel pedicellorum inflorescentiæ, globosi vel elongati atque turbinati, 2-5mm. longi, 0.5–2mm. diam., cooperti pseudo-membrana rosea quæ tandem contritur, expositis sporarum massis olivaceo-nigris atque agglutinatis. Sporæ castaneo-brunneæ, sphaericæ, globosæ, late ovatæ, lateraliter compressæ, diam. 13—18μ medietate 15μ; episporium crassissimum, ferrugineum, leve ; quæ in sporis continentur magnapere granosa.

Habitat Cyperi sp. Cocanada, Madras ; leg. S. Sundararaman, decembri, 1906.

LVIII **Farysia americana** Ciferri, *Ann. Mycol. Berl.* **29** : 73, 1931.

Ovariicolous ; all ovaries destroyed, sori 3-6mm. long, globular to elongate, covered by whitish papery pseudo-membrane ; membrane soon flakes away exposing the sooty-black (Ridgway) spore-mass, with a columella at base and strands of hyphæ (elators) traversing the sorus. Spores cinnamon-brown (Ridgway), spherical, globose to subglobose, slightly angular, occasionally elongate, 4-9μ in diameter with a mean of 6 ; epispore thick and punctate.

On *Carex baccans* Nees at Kodaikanal ; collected by T. S. Ramakrishnan on December 6, 1944.

This species comes nearest to *Farysia americana* Cifferi agreeing in all respects, excepting that the spores of the type are 4—10μ with a mean of 6μ, while those of Kodaikanal collection are 4—9μ with a mean of 6μ.

LIX **Sorosporium digitariæ** (Kunze) Padwick, *Paper, Imp. Mycol. Inst.* **17** : 6—8, 1946.

Syn. *Ustilago digitariæ* (Kunze) Winter, *Rabenh. Krypt. Fl.* **1** : 88, 1881 ; Saccardo, *Syll. Fung.* **7** : 454, 1888 : Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India,* **1** : 48, 1931.

Sphacelotheca digitariæ (Kunze) Clinton *N. Amer. Fl.* 7 : 998, 1939.

Sori infecting the inflorescence, developing into dark Quaker-drab (Ridgway), elongate bodies protruding out of leaf-sheaths, 2—5cm. long, covered at first by a pinkish papery pseudo-membrane ; membrane later flakes away exposing the blackish-brown, dusty spore-mass and numerous filiform shreds ; sterile cells cubic or elongate, hyaline, verrucose, thick-walled, having about the same size as spores. Spore-balls elliptical or spherical ; spores cinnamon-brown to chestnut-brown (Ridgway), globose to subglobose, 5.5—7.5μ in diameter with a mean of 6.5μ ; epispore medium, thick, smooth, with radial striations.

On *Urochloa reptans* Stapf (= *Panicum repens* Linn.) at Harley, S. Mysore, Collected by C.A. Barber on September 15, 1908 ; at Hunsur, Mysore (E. J. Butler on September 21, 1903) ; at Yelwal, Mysore (E. J. Butler on September 25, 1903) ; at Tejgaon, Dacca, Bengal (G. W. Padwick on May 21, 1945).

LX **Sphacelotheca apludæ** sp. nov.

Syn. *Ustilago arundinellæ* Sydow and Butler, *Ann. Mycol. Berl.* **5** : 484, 1907, (*not* Brefeld); Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India.* **1** : 48, 1931.

Sori infecting all the ovaries and destroying them completely ; forming purplish-grey elongate bodies covered by glumes, 5—7.5mm. long, with a leathery peridium enclosing a powdery spore mass and a long persistant columella. Columella 1.5—2cm. long, protruding out, slender and forked at the tip, forked ends sometimes curved. Spores Buckthorn brown (Ridgway), irregularly globose, elliptical, conical with blunt angles, 5.5—8μ in diameter with a mean of 7μ ; epispore thin and smooth.

On *Apluda varia* var. *aristata* Hack. at Kumaon, Himalayas. Collected by E. J. Butler on June 24, 1907.

This grass was identified as *Arundinella setosa* Trin. and the smut on it identified by Butler and Bisby as *Ustilago arundinellae* Brefeld, both of which are misdeterminations.

Sori ovaria omnia inficientes atque penitus destruentes, purpureogrisea corpora cooperta glumias efformantes, 5—7.5mm. longi, coriaceo peridio ornati includente pulverulentam sporarum massam columella persistentes. Columella 1.5—2cm. longa, eminenter prominens, tenuis, bifida in apice, duplici brachio nonnumquam curvato. Sporæ 'Buckthorn brown' (Ridgway) irregulariter globosae ellipticæ, turbinatæ angulis obtusis ; diam. 5.5—8μ medietate 7μ ; episporium tenue, leve.

Habitat Apluda varia var. aristata Hack. Kumaon, U. P.; leg. E. J. Butler, 24-6-1907.

LXI **Sphacelotheca aristidæ-cyananthæ** (Brefeld) comb. nov.

Syn. *Ustilago aristidæ-cyananthæ* Brefeld, *Unters Gesammt. Mykol.* **12**: 102, 1895; Saccardo, *Syll. Fung.* **14**: 415, 1899; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India.* **1**: 47, 1941.

Sori infecting the ovaries, forming olive-brown to deep-olive (Ridgway) cylindrical bodies, 0.5—2.5cm. long and 1—2mm. broad, having a tough leathery wall and enclosing a blackish-brown, compact spore-mass and a delicate central columella. Sterile cells hyaline, spherical and smooth. Spores Prout's brown (Ridgway), globose to subglobose or polygonal, 6—10μ in diameter with a mean of 8μ; epispore slightly dark and finely echinulate.

On *Aristida* sp. at Dehra Dun. Collected by R. .S. Hole on December 27, 1908.

Compared with the type specimen kindly sent by Dr. E. Ulbrich, Curator, Berlin Botanical Museum and found to agree very well.

LXII **Sphacelotheca cypericola** sp. nov.

Ovariicolous; sori minute, infection scattered; sori less than half mm. in dimensions, black, with a tough leathery peridium enclosing brownish spore-mass and a stout central columella. Spores Sayal brown to Tawny olive (Ridgway), spherical, globoid, occasionally elongate or oval, 7—11μ in diameter with a mean of 9μ; epispore thin and reticulate.

On *Cyperus difformis* Linn. at Ladhar. Collected by S. Ahmad on November 23, 1935.

This appears to be the first reord of a *Sphacelotheca* on *Cyperus*. The only other *Sphacelotheca* on Cyperaceæ is *Sphacelotheca caricis-petitianæ* Zundel.

Ovariicola; sori minuti, infectio dispersa; sori dimidio millimetro minores, nigri, duro coriaceo peridio ornati, bunneas sporarum massas includentes columella centrali crassa. Sporæ 'Sayal brown' ad 'Tawny olive' (Ridgway), sphæricæ, globoideæ, nonnumquam elongate vel ovatæ, diam. 7—11μ, medietate 9μ; episporium tennen reticulatum.

Habitat Cypero difformi Linn. Ladhar, Punjab, leg.; S. Ahmad, 23-11-1935.

LXIII **Sphacelotheca destruens** (Schlecht.) Stevenson and Johnson, *Phytopathology*, **34**: 613, 1944.

Syn. *Cœoma destruens* Schlecht., *Fl. Berol.* **2**: 130, 1824; Link in *Willdenow* Sp. *Plant.* **6**: 3, 1824.

Tilletia destruens Lev., *Ann. Sci. Nat. Bot.* **3**, **8**: 372, 1848.

Ustilago panici-miliacei Wint., *Rabenh. Krypt. Fl.* **1**: 89, 1884. Saccardo, *Syll. Fung.* **7**: 454, 1888; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1**: 50, 1931.

Sorosporium panici-miliacei Takahashi, *Bot. Mag. Tokyo*, **16**: 184, 247, 1902.

Sphacelotheca panici-miliacei Bub., *Houby Ceske*, **2**: 27, 1912.

Sori developing in the inflorescence and completely destroying it, forming a flask-shaped body, 4 x 2cm. in dimension, with a pale yellow leathery pseudo-membrane, enclosing the blackish brown, compact spore-mass. Spores in balls, Mummy-brown (Ridgway) subspherical to globose, ovate, 5.5—9μ in diameter with a mean of 8μ ; epispore medium thick, smooth and tuberculate.

On *Panicum miliaceum* Linn. at Larkipur, Kashmir. Collected by E. J. Butler on September 11, 1908.

This smut appears to belong to the genus *Sorosporium* due to the presence of evanescent spore-balls but an examination of the type specimen is necessary before making the transfer.

LXIV **Sphacelotheca erythraænsis** (Sydow) Clinton, *N. Amer. Fl.* **7** : 996, 1939.

Syn. Ustilago erythraœnsis Sydow, *Ann. Mycol. Berl.* **9** : 144, 1911 ; Saccardo, *Syll. Fung.* **23** : 611, 1925 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India.* **1** : 43, 1931.

Ovariicolous ; sori developing deep Quaker-drab (Ridgway), elongate bodies, 5—10mm. long with a leathery peridium containing sooty-black (Ridgway) spore-mass and a persistent columella. Sterile cells few and scattered. Spores Prout's brown to olive-brown (Ridgway), spherical, globose to broadly oval, 8.5—12μ in diameter with a mean of 10.5μ ; epispore thick and aculeate.

On *Manisuris* sp. at Dharwar. Collected by E. J. Butler on September 23, 1912 ; at Dohad, Bombay (E. J.Butler on October 9, 1912) ; on *Manisuris granularis* Linn. at Pusa. Collectd by E. J. Butler on September 28, 1907 and A. Khan in September, 1936 ; at Jaitpur, Nepal (I. H. Burkill on December 8, 1907) ; at Belari, Amaraoti (I. H. Burkill on October 6, 1908).

LXV **Sphacelotheca monilifera** (Ellis and Everhart) Clinton, *J. Mycol.* **8** : 141, 1902.

Syn. *Ustilago monilifera* Ellis and Everhart, *Bull. Torrey Bot. Cl.* **22** : 362, 1895 ; Saccardo, *Syll. Fung.* **14** : 420, 1899.

Sori ovariicolous ; forming dark-drab (Ridgway) elongate bodies, covered by glumes, 6—7mm. long, with a pinkish pseudo-membrane which ruptures later on disclosing blackish-brown dusty spore-mass with a persistent columella. Spores Mummy-brown (Ridgway) globose to subglobose, broadly oval, 8.5—13μ in diameter with a mean of 11μ ; epispore thick, ferruginous (Ridgway) and aculeate.

On *Heteropogon contortus* Beauv. (= *Andropogon contortus* Linn.) at Walyar, Palghat, S. Malbar. Collected by the Govt. Mycologist on January 18, 1924 in North Malbar ; in Nandi Hills, Mysore (M. J. Thirumalachar on November 18, 1944).

LXVI **Sphacelotheca tenuis** (Sydow) Zundel, *Mycologia*, **22** : 137, 1930.

Syn. *Ustilago tenuis* Sydow, *Ann. Mycol. Berl.* **4** : 425, 1906 ; Saccardo, *Syll. Fung.* **21** : 506, 1912 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India.* **1** : 50, 1931.

Sori infecting the ovaries and covered by glumes ; forming purple-grey (Ridgway) cylindrical bodies, 5—10mm. long, enclosing blackish-brown powdery spore-mass with a persistent columella. Spores chestnut-brown (Ridgway), spherical, globose to subglobose, broadly oval, rivulose, 5.5—10μ in diameter with a mean of 7.5μ ; epispore thick, ferruginous (Ridgway) and smooth.

On *Bothriochloa pertusa* A. Camus (= *Andropogon pertusus* Linn.) at Palamkotta, Madras. Collected by C. A. Barber on May 11, 1903 ; at Hunsur, Mysore (E. J. Butler on September 21, 1903).

LXVII **Sphacelotheca tonglinensis** (Tracy and Earle) Zundel, *Mycologia*, **36** : 406, 1944.

Syn. *Ustilago tonglinensis* Tracy and Earle, *Bull. Torrey Bot. Cl.* **22** : 175, 1895 : Saccardo, *Syll. Fung.* **14** : 250, 1899 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, 1 : 50, 1931.

Sori ovariicolous ; developing into elongate purplish-gray bodies, half-covered by outer glumes, with a tough leathery pseudo-membrane, containing blackish-brown spore-mass with a persistent columella. Spores carob-brown (Ridgway), spherical, broadly oval, 8—13μ in diameter with a mean of 10μ ; epispore thick, ferruginous (Ridgway) and aculeate.

On *Ischæmum* sp. at Chatrapur, Ganjam, Orissa. Collected by C. E. C. Fischer in October, 1904 ; at Hoshangabad, C. P. (E. J. Butler on September 29, 1912) ; at Samalkot, Madras (F. J. F. Shaw on November 21, 1910) ; on *Ischæmum spathiflorum* Hook. f. at Girnar, Junagad (G. S. Kulkarni on September 23, 1916).

The fungus was incorrectly recorded by Butler and Bisby as *Ustilago burmanica* Sydow and Butler. Re-examination shows that it is *Sphacelotheca tonglinensis* (Tracy and Earle) Zundel.

LXVIII **Tilletia ahmadiana** sp. nov.

Ovariicolous ; sori completely destroying the ovaries, 0.5—1.5mm. long, covered by the outer glumes, containing dusty black spore-mass bounded by the ovary walls ; sterile cells hyaline, thick-walled, warty, almost the same size as spores. Spores Diamine-brown (Ridgway) spherical, broadly oval, 16.5—20.5μ in diameter with a mean of 18.5μ ; epispore thick, with papillate warts.

On *Perotis latifolia* Ait. at Jaggatpur, Gurdaspur. Collected by S. Ahmad on September 18, 1941.

Ovaricola ; sori penitus ovaria destruentes, 0.5—1.5mm. longi, operti glumi externis, continentes pulverulentam sporarum nigram massam circumscriptam ovarii parietibus. Cellulæ steriles hyalinæ, crassis parietibus præditæ, verucosæ, magnitudine fere eadem ac sporæ. Sporæ 'Diamine brown' (Ridgway) sphæericæ, late ovatæ, diam. 16.5—20.5μ, medietate 18.5μ ; episporium crassum papillatis verrucis orantum.

Habitat Peroti latifolia Ait ; Jaggatpur, Gurdaspur, Punjab. Leg. S. Ahmad, 18-9-1941.

LXIX **Ustilago crameri** Koernicke in Fuckel, *Symbol. Mycol. Nachtrag.* **2** : 11, 1873 ; Saccardo, *Syll. Fung.* **7** : 455 1888 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 48, 1931.

Sori infecting the ovaries, inner glumes and main rachis ; blackish-brown spore-mass enclosed by papery shreds of the outer glumes ; sterile cells smaller than spores, spherical and hyaline. Spores Buckthorn-brown (Ridgway), globose to subglobose, oval, 6.5—11μ in diameter with a mean of 8.5μ ; epispore thick, smooth and punctate.

On *Setaria italica* Beauv. at Niphad, Dist. Nasik. Collected by the Agricultural Officer in charge Cereal Breeding Station on November 6, 1943; at Haveri, Dharwar (G. S. Kulkarni in October, 1915) ; at Hyderabad Dn. (Agricultural Chemist, H.E. H. Nizam's Govt. in May, 1942) ; at Manjri, Poona (H. M. Chibber on October 25, 1908) ; at Poona (G. S. Kulkarni on September 30, 1910).

LXX **Ustilago cynodontis** P. Hennings, *Engl. Bot. Jahrb.* **14** : 369, 1831 ; Saccardo, *Syll. Fung.* **14** : 416, 1899 : Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India.* **1** : 49, 1931.

Sori infecting the spikelets, main rachis and leaf-sheaths, latter turning into grey-coloured shreds by distortion ; sori at first enclosed in the leaf-sheaths, opening later on to expose blackish-green-grey (Ridgway), powdery spore-mass ; sori developing on leaf-sheaths in advanced stages of infection. Spores Buckthorn-brown to Prout's brown (Ridgway), spherical to globose, oval, 5.5—9μ in diameter with a mean of 7μ ; epispore thick and smooth.

On *Cynodon dactylon* Pers. all over India wherever the host is found.

LXXI **Ustilago effusa** Sydow, *Ann. Mycol. Berl.* **4** : 425, 1906 ; Saccardo, *Syll. Fung.* **21** : 506, 1912 : Butler and Bisby, *Sci. Monogr. Coun. Agric. Res., India*, **1** : 49, 1931.

Foliicolous ; sori on the ventral surface of the leaf ; youngest rolled leaf usually attacked, on unrolling exposes the dusty, olive-brown spore-mass ; sterile cells hyaline, spherical, verrucose and thick-walled. Spores Mummy-bown (Ridgway) spherical, globose to subglobose, 3.5—7.5μ in diameter with a mean of 5μ ; epispore dark, thick and echinulate.

On *Arundinella wallichi* Nees at Wahjan, Assam. Collected by E. J. Butler on May 16, 1905 ; on *Chrysopogon aciculatus* Trin. (= *Andropogon aciculatus* Retz.) at Cherrapunji, Assam (L. S. S. Mony on October 27, 1914) ; on *Vetiveria ziganoides* Stapf (= *Andropogon muricatus* Retz.) at Kanaighat, Sylhet (E. J. Butler on May 21, 1905).

LXXII **Ustilago egenula** Sydow and Butler, *Ann. Mycol. Berl.* **10** : 251, 1912 ; Saccardo, *Syll. Fung.* **23** : 609, 1925 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 49, 1931.

Ovariicolous ; sori forming hypertrophied, conical, tough, olive-coloured bodies ; half-covered by the glumes ; infection scattered in the panicle. Spores Mummy-brown (Ridgway), dusty, spherical to globose, broadly oval, 7.5—13μ in diameter with a mean of 10.5μ ; epispore rather thick, ferruginous (Ridgway) and aculeate.

On *Eragrostis nutans* Nees at Pusa. Collected by E. J. Butler on December 8, 1910 ; on *Eragrostis japonica* Trin. at Ladhar, Punjab (S. Ahmad).

LXXIII **Ustilago indica** Sydow and Butler, *Ann. Mycol. Berl.* **10** : 250, 1912 ; Saccardo, *Syll. Fung.* **23** : 610, 1925 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 49, 1931.

Sori in culms, altering the floral axis into a long, sometimes curved fragile body, 2—4cm. long, 1—1.5mm. broad, covered by a thin silvery-grey membrane formed of host tissue ; membrane flakes away at maturity. Spores Prout's brown (Ridgway), irregularly globose, ovate to elongate, occasionally with a depression on one side, 5.5—8.5μ in diameter with a mean of 6.5μ ; epispore dark, thick and punctate.

On *Eulaliopsis binata* (Ritz). C. E. Hubbard at Pathankot, Punjab. Collected by J. H. Mitter on May 22, 1911.

This grass was named *Ischœmum angustifolum* Hack. = *Pollinia eriopoda* Hance by Sydow and Butler and also Butler and Bisby, which is a misdetermination.

LXXIV **Ustilago maydis** (De Candolle) Corda, *Icones Fung.* **5** : 3, 1842

Syn. *Lycoperdon Zeœ* Beckman,, *Hannov. Mag.* **6** : 1330, 1768.

Uredo segatum var. *mays-zeœ* De Candolle, *Fl. France*, **2** : 596, 1805.

Uredo maydis De Candolle, *Fl. France*, **6** : 77, 1815.

Ustilago zeœ Unger, *Einfl. Bodens*, p. 211, 1836 ; Saccardo, *Syll. Fung.*, **7** : 472, 1888; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 51, 1931.

Ustilago schweinitzii Tulasne, *Ann. Sci., Nat. Bot.* 3, **7** : 86, 1847.

Ustilago mays-Zeœ Magnus, *Verh. Bot. Ver. Brandenb.* **37** : 7, 1895.

Sori developing in the ovaries causing hypertrophy, infection in tussels destroying the floral parts ; dusty brown spore-mass enclosed by a peridium of host-tissue and fungal hyphæ. Spores Mummy brown (Ridgway) globose to subglobose, ellipsoidal, 7.5—10μ in diametr with a mean of 9μ ; epispore dark, medium thick and bluntly echinulate.

On *Zea mays* Linn. at Srinagar, Kashmir. Collectd by E. J. Butler on August 3, 1908 ; at Harwan and Pampore, Kashmir (in August, 1908) ; at Munsung, Bengal (W. McRae on July 23, 1928 ; infection in tassel.)

Mundkur (1945) cited *Ustilago mays-zeœ* (DC) Corda as the correct name of this smut, which is an error.

LXXV **Ustilago microchloæ** Sydow and Butler, *Ann. Mycol. Berl.* **4** : 427, 1906 : Saccardo, *Syll. Fung.* **21** : 500, 1912 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 49, 1931.

Sori infecting the inflorescence, forming pyriform to globoid bodies, 1—3 x 2mm., bounded by a papery membrane of host-tissue and containing spores, blackish-green-

grey (Ridgway) spherical, broadly oval, 7.5—11μ in diameter with a mean of 10μ ; epispore thick, ferruginous (Ridgway), echinulate and punctate.

On *Microchloa setacea* Br. at Bilikere, Mysore. Collected by E. J. Butler on September 21, 1903.

LXXVI **Ustilago nuda** (Jensen) Rostrup, *Tidsskr. Landokonomi*, **8** : 745, 1889 : Saccardo, *Syll. Fung.* **9** : 283, 1891 : Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 49, 1931 : Mundkur, *Sci. Monogr. Coun. Agric. Res. India*, p. **12** : 12, 1938.

Sori infecting all spikelets in the ear destroying the floral parts, 5—10mm. long and 5mm. broad, covered by a thin papery pseudo-membrane ; membrane later flakes away exposing dark raw-amber (Ridgway) powdery spore-mass. Spores Prout's brown (Ridgway) globose, oval to ovate, darker on one side, 5.5—7.5μ in diameter with a mean of 6.5μ ; epispore medium thick and finely echinulate.

On *Hordeum vulgare* Linn. at all places in India where barley is grown.

LXXVII **Ustilago operata** Sydow and Butler, *Ann. Mycol. Berl.* **4** : 426, 1906 : Saccardo, *Syll. Fung.* **21** : 502, 1912 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 50, 1931

Sori destroying the ovaries ; half-covered in the outer glumes, enclosing dusty spore-mass and a conical, short, persistent columella. Spores Natal Brown to Verona brown (Ridgway), spherical to globose, broadly oval, 7.5—13μ in diameter with a mean of 10.5μ ; epispore thick, aculeate and tuberculate.

On *Brachiaria villosa* (Lamk.) A. Camus at Ootackmund, Nilgiri Hills at an altitude of 7500 ft. Collected by C. A. Barber on October 1, 1901 : at Nilgiris (?) (R. Proudlock on November 16, 1906) ; on *Brachiaria semiundulata* Stapf at Tukwar, Darjeeling, (A. Hafiz Khan on August 19, 1909).

LXXVIII **Ustilago panici-glauci** (Wallroth) Winter, *Rabenh. Krypt. Fl.* **1**: 97, 1881 ; Saccardo, *Syll. Fung.* **7** : 472, 1888 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 50, 1931.

Sori destroying ovaries and floral parts except the outer glumes, forming conical bodies containing a blackish-brown powdery spore-mass. Spores cinnamon-brown to Saccardo's umber (Ridgway), spherical, broadly oval, 7.5—11μ in diameter with a mean of 10μ ; epispore thick, ferruginous (Ridgway), aculeate and punctate.

On *Setaria glauca* Beauv. at Chaubattia, U. P. Collected by U. B. Singh on September 26, 1942 ; under the edge of Chikalda Plateau (I. H. Burkill on October 7 1908).

LXXIX **Ustilago rabenhorstiana** Kühn, *Hedwigia*, **15** : 4, 1876 Saccardo, *Syll. Fung.* **7** : 471, 1888 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 50, 1931.

Sori destroying spikelets, turning the main rachis of panicle into shreds, enclosed by leaf-sheaths, with blackish-brown powdery spore-mass. Spores Prout's brown to cinnamon brown (Ridgway), spherical, ovate to broadly oval, 8.5—13μ in diameter with a mean of 11μ ; epispore thick, ferruginous (Ridgway), aculeate and punctate.

On *Digitaria bifasciculata* (Trin.) Henr. at Simla. Collected by G. W. Padwick on September 5, 1941 ; on *Paspalum sangulinale* Lamk. at Tukwar, Darjeeling (A. Hafiz Khan on August 19, 1909).

LXXX **Ustilago royleani** Sydow and Butler, *Ann. Mycol. Berl.* **4** : 426, 1906 ; Saccardo, *Syll. Fung.* **21** : 499, 1912 ; Butler and Bisby, *Sci. Mongor. Coun. Agric. Res. India*, **1** : 50, 1931.

Sori infecting rachis of the spike and deforming it, blackish-brown powdery spore-mass borne naked. Spores Prout's brown (Ridgway), spherical to globose, broadly oval, tuberculate, 9.5—14μ in diameter with a mean of 11.5μ ; epispore thick, ferruginous (Ridgway) and aculeate.

On *Paspalum royleanum* Nees at Dehra Dun. Collected by E. J. Butler on October 10, 1903.

LXXXI **Ustilago shiraiana** P. Hennings, *Bot. Jahrb.* **28** : 260, 1900 ; Saccardo, *Syll. Fung.* **16** : 369, 1902 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 50, 1931

Sori on young twigs, 1—2mm., subepidermal, scattered, elongate and coalescent ; epidermis breaks off exposing olivaceous-black, powdery spore-mass. Spores Brussel's brown to Prout's brown (Ridgway), globose to subglobose, spindle-shaped on side-view, 6.5—11μ in diameter with a mean of 9μ ; epispore thin, smooth, occasionally rivulose.

On *Bambusa* sp. at Dehra Dun. Collected by A. Khan on May 22, 1904 ; at Poona (H. M. Chibber on September 20, 1908) ; at Gorakhpur, U.P. (G. W. Padwick on December 19, 1941).

LXXXII **Ustilago spermophora** Berkeley and Curtis, *N. Amer. Fungi, first series*, No. 1098 ; Saccardo, *Syll. Fung.* **7** : 466, 1888 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 50, 1931.

Sori infecting the ovaries ; ovaries slightly hypertrophied, enclosing a blackish-brown spore-mass. Spores cinnamon-brown to Saccardo's umber (Ridgway), spherical, broadly oval, 7.5—11μ in diameter with a mean of 10μ ; epispore thick, aculeate and tuberculate.

On *Eragrostis rhachitricha* Hochst. at Pusa. Collected by E. J. Butler on December 7, 1906 ; on *Eragrostis egentula* Steud, at Pusa (L. S. S. Mony on November 19, 1917) ; on *Eragrostis minoris* Host at Pathankot, Punjab (S. Ahmad).

LXXXIII **Ustilago trichophora** (Link) Kunze, *Flora*, p. 369, 1830 ; Koernicke, *Hedwigia*, **16** : 36, 1877 ; Saccardo, *Syll. Fung.* **7** : 462, 1888 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India*, **1** : 51, 1931.

Sori infecting ovaries ; ovaries slightly hypertrophied, half-enclosed by the outer glumes, containing olivaceous, powdery spore-mass. Spores chestnut brown (Ridgway), spherical, broadly oval, 6.5—13μ in diameter with a mean of 9.5μ ; epispore medium thick, ferruginous (Ridgway), aculeate and punctate.

On *Echinochloa colonum* Link at Dacca. Collected by P. Maheswari in August, 1940 ; at Yedehalli, Mysore (E. J. Butler on September 15, 1903) ; at Poona (E. J. Butler on August 25, 1903) ; at Nagpur (P. A. Pandit on August 20, 1908).

LXXXIV **Ustilago tritici** (Persoon) Rostrup, *Overs. K. Danske Vid. Salsk. Forh.* p. 15; 1890; Saccardo, *Syll. Fung.* **9**: 283, 1891; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India,* **1** : 51, 1931.

Sori infecting spikelets in the ear destroying all the floral parts, 5—10mm. long, 1—2mm. broad, elongate, confluent, half-covered by the leaf-sheath, exposing blackish-grey, loosely adherent spore-mass. Spores Prout's brown to cinnamon-brown (Ridgway), globose to subglobose, elliptical, 5.5—7.5μ in diameter with a mean of 6.5μ ; epispore medium thick and minutely echinulate.

On *Triticum vulgare* Vill. in every part of India where wheat is grown.

LXXXV **Ustilago tuberculiformis** Sydow, *Ann. Mycol. Berl.* **10** : 248, 1912 ; Saccardo, *Syll. Fung.* **17** : 473, 1905 ; Butler and Bisby, *Sci. Monogr. Coun. Agric. Res. India,* **1** : 51, 1931.

Sori attacking flowers, destroying floral parts ; dusky-brown powdery spore-mass enclosed by bracts and later exposed. Spores Natal brown (Ridgway) spherical, broadly oval, 10—13μ in diameter with a mean of 11.5μ ; epispore thick, ferruginous (Ridgway) and echinulate.

On *Polygonum chinense* Linn. at Darjeeling. Collected by W. McRae on July 19, 1909.

LXXXVI **Ustilago utriculosa** (Nees) Tulsane, *Ann. Sci. Nat. Bot.* 3, **7** : 102, 1847 : Saccardo, *Syll. Fung.* **7** : 476, 1888 ; Butler and Bisby, *Sci Monogr. Coun. Agric. Res. India,* **1** : 51, 1931.

Sori infecting the ovaries and the inflorescence, forming irregular bodies with host-tissue, enclosing Taupe brown (Ridgway) spore-mass. Spores Avellaneous brown to Wood brown (Ridgway) spherical to globose, broadly oval, 7.5—10μ in diameter with a mean of 9.5μ ; epispore medium thick and reticulate.

On *Polygonum orientale* Linn. at Lakhipur, Cachar, Assam. Collected by S. Chowdhury on March 3, 1942.

We wish to express our gratitude to Rev. Father H. Santapau S. J., of the St. Xavier's College, Bombay, for his kindness in rendering into latin the descriptions of new species.

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STUDIES ON THE FORMATION AND GERMINATION OF TELIOSPORES OF RUSTS I

By Raghubir Prasada

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GERMINATION of the teliospores of rusts with a long rest period is generally difficult to obtain. Unless they pass through the usual dormant period, little can be done, normally, to germinate them. Freezing, and alternate wetting and drying may help in breaking the dormancy to a certain extent but the precise temperatures that would promote germination have also to be determined to get good results.

Among such teliospores, those of *Puccinia graminis* Pers. var. *tritici* Erikss. & Henn., *Puccinia triticina* Erikss. and *Puccinia glumarum* (Schm.) Erikss. & Henn. which attack wheat, are the most difficult to germinate. Not only have they been reported to have a long rest period but their germination and viability depends on the conditions that were prevailing at the time of and subsequent to their formation on their hosts.

This problem of formation of teliospores and their germination has been studied by the writer over a period of several years not only in the case of the above rusts but also *Puccinia penniseti* Zimm. on Bajra, *Pennisetum typhoides* Stapf & Hubbard and *Puccinia purpurea* Cke on Jowar, *Sorghum vulgare* Pers. It may be mentioned that in India the teliospores of *Puccinia graminis tritici* were first germinated by Mehta (1933). In a latter publication, he (1940) gave an account of the germination of *Puccinia triticina* also.

I PUCCINIA GRAMINIS *var.* TRITICI

The black rust of wheat is very common in the plains as well as the hills. In the Indo-Gangetic plain the telial stage appears in March-April and in the hills in April-May. A culture of this rust in the uredial stage has been maintained in the miniature plots at Simla all the year round for several years and the germination of teliospores formed therein tested from time to time. Collections made from the crop at Simla and some places in the plains were also tested on several occasions. In addition, teliospores of Indian physiologic races were raised in the greenhouse by inoculating adult plants of a susceptible variety. It was found that races 24, 34, 40 and 42 formed telia more or less readily when the host plants reached maturity, whereas races 15, 21 and 75 did so only rarely and then after a long period. Garbowski (1939) also had a similar experience with races 34, 40 and 21.

Lambert (1929), Cotter (1932), Sibilia (1930) and Johnson (1941) found that material kept dry and at a low temperature remained viable for the longest time. Johnson (1941) was able to preserve the viability of teliospores of race 9 for six years and 2½ months by storing them at 8°—10°C. preceded by freezing for 7½ months.

Several times during the course of these studies teliospores were kept in ice cubes in a refrigerator after the spores had been brought to germinable condition by breaking their dormancy through artificial freezing followed by alternate drying and wetting. At the end of two years, the material gave nearly 25 per cent germination as compared to 60 per cent to start with. Material stored in the room at 10°–20° C. simultaneously, lost all viability after 6½ months.

Johnson (1931) reduced the dormancy of teliospores to 20 days by simulating natural conditions to which they are exposed. By adopting the same methods, germination of teliospores formed in miniature plots early in winter at Simla was secured by the writer within 30 days of their formation.

It was found necessary to freeze the material before teliospores would germinate because collections as old as one year did not yield to alternate wetting and drying, or prolonged wetting alone.

Once the dormancy had been broken by freezing and alternate wetting and drying, there was no appreciable difference in germination with longer storage. Alternate wetting and drying was found to improve and also reduce the time taken for germination. After the infected straw had been soaked overnight in water the minimum time taken by the teliospores to germinate was found to be 8 hours.

(a) Temperature at the time of formation

Johnson (1931) stated that teliospores formed at 10°—16°C. germinate more abundantly and consistently than those produced at 21°—24°C. During these studies teliospores collected from the wheat crop at Agra, in the plains, during March and at the lower altitudes of Simla hills (3,000—5,000ft. a. s. l.) in May—June were not found to be viable. Out of collections made from the crop at Simla in May, occasional germination was noticed only in those obtained from the shade. Teliospores collected from fields exposed to the sun throughout the day failed to germinate.

Considering that maximum shade temperature in Simla during May often touches 30° C. it is evident that teliospores formed at that temperature possess very little viability, if at all. Those produced in the sun when the shade temperature is 30° C. do not germinate even after artificial treatments.

Teliospores collected from Narkunda (9,200ft. altitude) after they had been exposed to winter snows gave nearly 40 per cent germination without further treatment. Similarly, teliospores formed in the miniature plots at Simla during winter gave 40—90 per cent germination.

(b) Effect of exposure to higher temperature after formation

The viability of teliospores not only depends on the temperature at the time of formation but also on subsequent exposure. Lambert (1929) observed that the viability of teliospores from Texas, Oklahoma, Southern Kansas, etc., that are formed during May—June is destroyed by exposure to hot summer in July-September. According to Cotter (1932) teliospores lose their viability within a week if exposed to natural conditions.

The effect of exposure to natural conditions at Simla was determined during these studies when viable teliospores were put in wire netting frames in the open on April 1, 1940 and 1941. The results given in Table I show that nearly all the teliospores were killed by the summer heat. Weekly average minimum and maximum as well as the lowest and highest temperatures in shade in April, May and June, 1940 and 1941, recorded at Simla, are given in the Appendix.

Teliospores were also exposed to different temperatures between 32° and 50° C. for 6 to 24 hours after which their viability was tested. It was found that exposure

to 32°—38° C. for 36—48 hours killed all the teliospores ; only 5—10 per cent germination was noticed after 12 hours' exposure at 38°—44°C whereas all the teliospores were killed after 24 hours at 38°—44° C. and 18 hours at 45°—50°C.

TABLE I

The effect of exposure of teliospores of Puccinia graminis *var.* tritici *to natural conditions and to conditions in a room at Simla, as compared to their storage at 5°—7° C, on their viability.*

Period of Exposure or Storage	Date of testing germination	Percentage germination of teliospores stored in		
		Open	Room	5°—7°C.
1940				
April 1—15	15-4-40	50	70	80
„ 1—30	30-4-40	30	50	80
„ 1—May 15	15-5-40	10	50	90
„ 1— „ 31	31-5-40	5	40	80
„ 1—June 15	15-6-40	0	30	75
„ 1— „ 30	30-6-40	0	25	80
1941				
April 1—15	15-4-41	40	60	80
„ 1—30	30-4-41	10	50	75
„ 1—May 15	15-5-41	T	40	80
„ 1— „ 30	30-5-41	0	33	90
„ 1—June 15	15-6-41	T	20	80
„ 1— „ 30	30-6-41	0	10	90

T =Trace, *i.e.*, less than 1 per cent

(c) Influence of temperature on germination

According to information available in literature, teliospores of *Puccinia graminis* germinate best and most consistently between 12°—18° C. As reported by Mehta (1940), satisfactory results were obtained at these temperatures in all the tests made in this country since 1933. During the course of these studies, slightly lower and higher temperatures were employed and it was found that germination of teliospores does not take place below 10° and above 20° C. but 12—18° C. was most suitable.

II PUCCINIA TRITICINA

The brown rust of wheat is common in the plains as well as the hills, but the telial stage is very scarce and often impossible to obtain in this country. In the Indo-Gangetic plain the teliospores sometimes appear in March—April ; once they were collected as early as December from Gorakhpur. As recorded by Mehta (1933, 1940) and observed by the writer for several years, the telial stage of this rust is not

formed on the crop at Simla. Butler (1918) observed that their formation depends on special climatic conditions. According to Mains (1924) wheat seedlings never produce telia in the greenhouse even on old drying leaves but some telia are formed on old leaves of mature plants. Waters (1928) found that subjecting the rusted plants to darkness, low temperatures, dessication, etc., did not bring about the formation of teliospores in *Puccinia triticina*. Montemartini (1939) observed that teliospores of *P. triticina* never develop normally under Sicilian conditions, as the infected plants dry up too soon ; when they do occur on the straw, they seldom germinate.

The presence of teliospores on drying self-sown plants has been sometimes observed during winter at Simla. This may be due to the gradual loss of water at low temperature. While no case of telial formation on wheat crop at Simla is known to the writer, it is interesting to report its formation, in abundance, on wheat variety IP 114. This variety was inoculated along with several others in the heading stage with a mixture of all the Indian physiologic races of brown rust in April. By the end of May all the leaves of IP 144 were found to contain a large number of telia whereas no other variety, resistant or susceptible, produced teliospores. It appears, therefore, that apart from Butler's observation that the formation of teliospores in *P. triticina* depends on special climatic conditions, other factors are also involved in the process. IP 114 is susceptible to brown rust and it is difficult to explain the formation of teliospores on this variety of wheat alone. Since this observation was made in 1940, IP 114 has been grown in miniature plots at Simla, inoculated with brown rust and teliospores obtained at will whenever required.

If stored dry at 5—7° C. the teliospores remain viable for nearly two years but lose their viability after one year in the room or exposed to natural conditions at Simla.

Mains (1924) observed that teliospores of *P. triticina* sometimes germinate without overwintering. During these studies teliospores formed at Simla on IP 114 either in summer or winter germinated soon after formation without any special treatment. In general, germination improved as the collections matured. As shown in Table II, an improvement in germination from 25 to 80 per cent was recorded after keeping the material for three months. It was found that teliospores stored in the room or exposed in the open at Simla showed an increase in germination for nearly six months after which there was a gradual decline. Alternate wetting and drying improved germination. Freezing was not found essential but made a decided improvement. The minimum time taken by the teliospores to germinate was 48 hours.

(a) Influence of temperature on germination

The precise range of temperature that is suitable for the germination of teliospores of *P. triticina* is not known. Mehta (1940) observed that temperature employed for the germination of teliospores of black rust were found to be satisfactory for teliospores of brown rust.

Tests made under natural conditions all the year round at Simla and at different temperatures show that teliospores of *P. triticina* can germinate between 7° and 27°C. and the optimum range is 10—16° C.

(b) Effect of temperature at the time of formation on the germinability of teliospores

In *Puccinia graminis var. tritici* it has been shown that teliospores formed on the crop which was exposed to the sun at Simla did not germinate and only those formed in the miniature plots at a low temperature during winter gave consistent and good germination; collections made from the crop in the plains also did not germinate. On the other hand, teliospores of *Puccinia triticina* collected from Agra in March, Gorakhpur in December and Simla in May germinated freely without any special treatment. All these observations indicate that teliospores of brown rust tolerate higher temperatures more than those of black at the time of their formation. Conditions in the sun when the shade temperature was 30°—32°C. proved to be lethal to teliospores of black rust but not to those of brown rust.

(c) Effect of exposure to higher temperatures after formation

According to Sibilia (1931) teliospores of *Puccinia triticina* tolerate high temperatures (up to 65° C for a few minutes) with no appreciable loss of germinating power. During these studies material collected from IP 114 on 25th May, 1940, which gave nearly 30 per cent germination soon after collection, was exposed to natural condition at Simla during June, July and August and its viability was compared with that stored in the room and at 5°—7° C. The results are given in Table II.

TABLE II

Germination of teliospores of Puccinia triticina *kept under different conditions at Simla*

Period of Exposure 1940	Percentage germination of material kept in		
	Open	Room	5°—7° C.
June 1—15	30	33	25
„ 1—30	40	40	50
„ 1—July 15	50	50	60
„ 1— „ 31	40	60	60
„ 1—Aug. 15	25	75	80
„ 1— „ 31	50	80	80

Results recorded in Table II show that teliospores of *Puccinia triticina* remain viable through the summer at Simla. As already shown teliospores of *P. graminis* var. *tritici* treated similarly were killed when exposed to natural conditions.

When viable teliospores of *P. triticina* were subjected to temperatures between 30° and 50°C. it was found that less than 10 per cent germination was shown by them after 48 hours at 38—44° C. and all the teliospores were killed after 24 hours at 45—50° C. The controls gave 60—80 per cent germination in every test.

III PUCCINIA GLUMARUM

The yellow rust of wheat occurs commonly in the plains of Northern India and the Nilgiri and Palni hills. The telia are formed commonly at all stages of growth of the host, sometimes within a fortnight of appearance of rust. In greenhouse cultures they are formed in about three weeks. Out of all the Indian physiologic races teliospores are produced very readily in race D and within ten days of appearance of rust. In yellow rust teliospores are formed in every season but more readily when the weather is warm and unfavourable for the propagation of the rust in the uredial stage.

Teliospores kept dry at 5—7°C. retained their viability for about a year. Stored in a room at Simla they remained viable for six months. No germination was obtained after the material had been exposed to natural conditions during April, May and June at Simla. It was found that temperatures between 10—20°C. are quite suitable for germination with optimum near 14—16°C. A period of 12 hours in the moist chamber was sufficient to bring about their germination under optimum conditions.

Teliospores collected at Simla from greenhouse cultures, miniature plots, the crop in March and volunteer plants in October have been found to germinate soon after collection without any special treatment. In race D nearly 90 per cent germination was obtained within ten days of their formation. Collections from the crop at several places in the plains received during March did not show any viability, indicating that teliospores are killed by heat prevailing in the plains at the time of their formation. All the viability is lost when teliospores are exposed to 38—40° C. for 24 hours.

4. PUCCINIA PENNISETI AND PUCCINIA PURPUREA

These rusts have been reported from several places in U.P., C.P., Bihar, Bombay and Madras (Butler & Bisby, 1931). Cultures were maintained in a greenhouse at Simla on Bajra and Jowar respectively and teliospores that were formed in all the seasons irrespective of the stage of growth of the hosts were tested for germination.

Temperature between 10° and 20° C. was found to be suitable for the germination of teliospores which do not require a period of rest. Exposure to 35—38°C. for 24 hours did not materially affect their viability but they were killed in 36 hours at 45—50°C.

It was found that teliospores retained their viability for 9 months if kept dry at 5—7° C. or in the room at Simla. Part of the same collection kept in a room at Agra during summer also retained its viability for 6—8 months. Older collections, however, germinated poorly and after longer soaking, which shows that teliospores of these rusts do not retain their viability for a long time. Teliospores started germinating within 12 hours in the moist chamber between 10—20° C.

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SUMMARY

The formation, storage and germination of teliospores of *Puccinia graminis* var. *tritici*, *Puccinia triticina*, *Puccinia glumarum*, *Puccinia penniseti* and *Puccinia purpurea* were studied.

In *P. glumarum*, *P. penniseti* and *P. purpurea* the uredial stage was invariably followed by the formation of teliospores, irrespective of the stage of growth of the host, while in *P. graminis* var. *tritici* teliospores were generally formed on mature plants and in *P. triticina* under special climatic conditions. Wheat variety IP 114 seems to possess some properties which induce the formation of teliospores in *P. triticina* freely in every season.

Stored dry and at a low temperature (5°—7°C.) the teliospores of all the rusts retained their viability for the longest period.

A rest period of 30 days was found only in the case of teliospores of *P. graminis* var. *tritici*. Teliospores of all other rusts germinated soon after formation.

Teliospores formed during the cold weather germinated abundantly and consistently. Their viability was adversely affected by exposure to high temperature after formation.

For the germination of teliospores temperatures between 12°—18° C. were found to be favourable and alternate wetting and drying proved to be beneficial in every case.

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APPENDIX

Weekly average minimum and maximum as well as the lowest and highest temperatures recorded in shade each week in the months of April, May and June during the years 1940 *and* 1941 *at Simla.*

Period	Weekly average °C. Min. °C.	Max. °C.	Lowest °C.	Highest °C.
1940				
April 1—7	11	17.5	9	19
„ 8—14	14	21	13	23
„ 15—21	12	18	10	21
„ 22—28	14	19	13	21
„ 29—May 5	15	20	13	21
May 6—12	17	22	15	24
„ 20—26	17.5	22	15	24
„ 27—June 2	17.5	23	17	25
June 3—9	16	21	14	24
„ 10—16	18	23	16	14
„ 17—23	16	21	15	23
„ 24—30	17	20	17	21
1941				
April 1—7	9	19	8	20.5
„ 8—14	12	23	11	24.5
„ 15—21	15	26	12.5	28
„ 22—28	17	28	14.5	31
„ 29—May 5	20	31	20	32.5
May 6—12	13	25	8	30
„ 13—19	13	20	9	24
„ 20—26	16	26	13.5	28.5
„ 27—June 2	18	28	12	30.5
June 3—9	13	19	12	21
„ 10—16	15	20	14	22.5
„ 24—30	17.5	23.5	13.5	27

In inoculation trials in the green-house, the organism was pathogenic to *D. gangeticum* only and failed to produce any symptoms on any of the other species inoculated.

The organism is quite distinct from *X. desmodii* on *D. diffusum* and is therefore named as a new species namely *Xanthomonas desmodii-gangeticii* Uppal, Patel and Moniz, sp. nov.

A technical description of the organism is given.

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STUDIES ON LENTIL RUST, UROMYCES FABAE (PERS.) DE BARY IN INDIA

By Raghubir Prasada and Upendra Nath Verma

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DURING 1945-46 and 1946-47 there was a severe out-break of rust, *Uromyces fabæ*, on lentil (*Lens esculenta* L.) at Delhi. The rust is common on leguminous crops in India and abroad. In India it is known to attack lentil, pea, sweet pea and broad bean as reported by Butler (1918) in the Indo-Gangetic plain.

The life history of this rust had not been studied in India and little information was available on the factors which caused the annual recurrence of this disease. The experiments reported in this paper were undertaken in order to elucidate some of these factors. Normally, lentil is sown in October—November and harvested in April. Pycnia and æcia of the rust, which is autoecious, are observed on the leaves and stems in February. Within a few days of their appearance the disease spreads rapidly but only æcia are found on the infected parts. Most of the infected leaves do not show any pycnia, indicating the occurrence of secondary æcia. In general, the spread of the rust takes place by means of æciospores, the uredia appearing very late in the season, quickly followed by telia. The teliospores are formed in the same sorus as the urediospores. In cases of severe attack, even the legumes are found to be infected. After the harvest there is, as a rule, no trace of either the host or the rust left in the field.

THE TELIAL STAGE

The uredial stage is followed by the formation of teliospores in the same sorus and on the same mycelium. In nature, the teliospores are generally formed when the host reaches maturity and environmental conditions are unfavourabe for the propagation of the rust in the uredial stage. The teliospores are the resting spores in rusts but they may or may not require a dormancy period, depending on the rust and the environmental conditions. With regard to the teliospores of *Uromyces fabæ* no information is available regarding the resting stage, viability under different environmental conditions and the precise temperatures that would permit their germination. Since this information is necessary to determine the role of teliospores in the annual rust outbreaks, samples were collected from the field in April and stored in sealed glass tubes at 3—5° C. 8—10° C. 14° C. and 17—18° C. A part of this collection was exposed to natural conditions and the rest kept indoors.

Soon after collection in April, infected leaves and bits of stem were soaked in water overnight and teliospores were exposed to temperature of 7—8° C. 12—14°C. 17—18°C. 20—22°C. 29°C. and 30—35°C. for testing their germination. Within a week about 25 percent germination was observed in the material exposed to temperatures varying from 12° to 22°C. Such tests were conducted every month throughout the year with similar results, except that better germination, in a shorter period of 2—3 days, was observed at 17—18° C. and the percentage of germination improved with the approach of winter. Tests made during January—February yielded better and more consistent results than those made earlier because environmental conditions were ideal for their germination at that time. Alternate wetting and drying improved and hastened the process.

BACTERIAL SOFT ROT OF MANGO IN BOMBAY

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SOFT rot of mango fruits (*Mangifera indica* L.) is quite common during the season (May-August) when certain species of bacteria make entry through wounds or lenticels and rot the fruits by dissolving the middle lamella by the enzyme pectinase excreted by these organisms. Since no soft rot of mango had been recorded previously, studies were undertaken to determine precisely the organisms responsible for it.

Isolation :—The border of sound and softened tissue of mango fruit was sterilised in 0.1 per cent mercuric-chloride solution and small pieces removed by means of a flamed scalpel and dropped in sterilised distilled water for a few minutes after which dilution plates using neutral beef agar were poured, inverted after solidification and incubated at30°C. Small dirty white colonies which began to appear after the third day, were transferred for further studies as described below :—

Morphology.—The bacterium is a rod with rounded ends, single, never in chains. In 96 hour old culture grown in the nutrient broth at 30°C, the size of the cells is 2.09 (1.8 to 2.43) x 0.59 (0.54 to 0.72) μ; actively motile ; grayish dirty white ; gram-negative ; non-capsulated ; not acid fast ; no spores or involution forms.

Cultural characters.—On nutrient agar slants, the growth is fair, smooth, shining, opalescent, with irregular margin, filiform, no change in the colour of the medium and no distinctive odour while on potato dextrose agar slants, growth is copious, smooth, glistening, opalescent with irregular margins, pale olive grey colour and no distinctive odour in 96 hours. In nutrient broth, a fairly cloudy growth with sediment appears after 7 days. After 10 days' growth, litmus is completely reduced with reddish tinge at the top, no sediment, no floccules and no pellicle. Plain milk started clearing after 3 days with heavy pellicle, sediment and floccules. The pathogen is a strict aerobe. Optimum temperature for growth is 25°C. maximum 45°C. and minimum 5°C. Thermal death point is about 52°C.

Biochemical reactions.—Liquefaction of gelatin begins on the second day and proceeds very rapidly. In tryptophane broth, hydrogen sulphide production is evident after 7 days. The Gore' method shows positive (feeble) reaction for indol. The nitrites are formed after five days while ammonia is not. In Cohn's medium, there was heavy whitish growth with sediment but no pellicle after 7 days. In Uschinsky's medium, the growth was very heavy, whitish, cloudy with sediment, with no or thin pellicle and no flourescence. In Koser's uric acid medium, growth is positive. Acetyl-methyl-carbinol is produced.

Carbohydrates.—Acid and gas production takes place in dextrose, xylose, lactose, sucrose, raffinose, salicin and mannitol. Pellicle is formed in dextrose, xylose, sucrose, raffinose but none in others. Starch is not hydrolysed. Growth is good and light greenish-white in Simmon's citrate medium. On potato cylinders, the growth is copious, shining, cylinder turning light brownish, not flowing but covering the entire surface in 5 days. Casein is not digested. The pathogen grows on media with pH ranging from 5.3 to 9.0, the best growth occurring at 7 pH.

Pathogenicity.—In repeated trials, it was found that the organism is pathogenic on the following hosts viz., carrot, cucumber, bhendi (*Hibiscus esculentus*), french bean, tomato, chillies, radish, banana, sweet potato, peas, beet, potato, cauliflower, brinjal, cowpea pods, mosambi (*Citrus sinensis*) and mango while it failed to infect elephant foot (*Amorphophallus campanulatus.*), apple, lemon and garlic.

Identity.—Since the monumental work of Jones (1901) on *Bacillus carotovorus*, many new species have been created and much research work on rot producing organisms done by several investigators, chief amongst whom may be mentioned Harding and Morse (1909), Smith (1920), Lacey (1926), Dowson (1941) and Waldee (1945). Jones (1901) reported that this organism failed to attack potato but later work of Smith (1920) and Lacey (1926) has conclusively shown that *Bacillus carotovorus* does rot potato, which fact is also borne out in the present investigation.

According to Dowson (1941), four species of *Bacterium* and one of *Pseudomonas* are capable of producing soft rot of plants in Great Britain. The pathogen under reference does not belong to *Pseudomonas* group as it is dirty white and not yellowish green in colour. Of the four species of *Bacterium* producing soft rot, only two, viz., *B. carotovorus* and *B. aroideæ* have a wide range of natural hosts. The latter species however, is quite distinct from the former as it does not produce gas from several sugars commonly attacked by *B. carotovorus* producing acid and gas.

Since the organism under reference produces soft rot in mango and mosambi besides 15 vegetables and since acid and gas are produced from several sugars, it seems quite certain that the pathogen is *Bact. carotovorus.*

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BANDED LEAF BLIGHT OF ARROWROOT, MARANTA ARUNDINACEA

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In July 1946, during the period of the South-west monsoon, widespread incidence of a banded leaf blight of arrowroot (*Maranta arundinacea*) was reported from Ancharakandy, near Cannanore, in North Malabar. The disease was stated to be rapidly spreading among the plants, causing chlorotic banding of the leaves with ultimate browning, and rotting of the foliage. High incidence and quick spread were closely associated with periods of heavy rainfall. With the clearing of the weather and the onset of dry conditions, the severity of the disease usually diminished. The same disease had been observed in 1947 and 1948, in the months of July to October, in the same locality.

Characteristic symptoms of the foliage blight are visible in young and old leaves. Infection commences as small water-soaked spots which under favourable conditions enlarge rapidly. Meanwhile slender aerial hyphæ extend quickly over the surface of both sides of the leaf. The extension of the spot in the leaf is not always continuous but presents a succession of zones. Transverse necrotic patches, light green to yellowish brown in colour separated either by healthy green or bands of light green areas are found. The over-all effect on an affected leaf is the apparent formation of irregular bands across the blade extending throughout the width or confined to one half. Sometimes these bands are whitish. Meanwhile the hyphæ have grown over the whole leaf and cobwebby extensions to neighbouring leaf blades are apparent. The infection does not spread much to the sheath or down to the shoot. In the rainy season the affected leaves finally turn brown and may be involved in a wet rot greatly accelerated by secondary saprophytic organisms. Owing to severe blighting the growth of the plants and the formation of the rhizomes are adversely affected. With age, hyphæ become buff coloured and minute velvety; sclerotioid bodies are seen dotted on the lower surface. In the months of July—August the fungus forms a whitish superficial growth over the healthy portions. Fructifications which are observed to be basidia and basidiospores, are seen in these growths. An examination of the pathogen showed it to be *Pellicularia filamentosa* (Pat.) Rogers [= *Corticium solani* (Prill. & Del.) Bourd. & Galz.].

Pellicularia filamentosa enjoys a wide distribution. Kotila (1947) reports a foliage blight of sugar beet caused by this parasite in the United States of America. An areolate leaf spot of *Citrus* sp. has been described by Stahel (1940) from Surinam. The causal organism is described as a new species—*Corticium areolatum* Stahel—by the author. But Rogers (1943) is of opinion that the pathogen is synonymous with *Pellicularia filamentosa*. Jauch (1947) has described the occurrence of stem necrosis and death of broad bean plants, caused by *Pellicularia filamentosa* from Argentina. The same fungus was isolated from three other hosts, viz., *Cicer arietinum* L., pine and *Iberis* sp. Butler (1918) has recorded a banded sclerotial disease of sugarcane caused by a sterile fungus and states that it might probably be *Hypochnus sasakii* Shirai. He has mentioned that the same fungus was known on *Oryza sativa* L., *Saccharum spontaneum* L., *Saccharum arundinaceum* Retz., *Rottboellia exaltata* L., and *Maranta sp.* and that it could pass on from these hosts to sugarcane. Bell (1929) reports that the causal agent of the banded sclerotial disease of sugarcane in Australia belongs to the *mycelia sterila* group. Subramaniam (1936) has named the pathogen on sugarcane as *Hypochnus sasakii*. But these authors had not seen the perfect

state. Ryker (1939) reported that sugarcane infected by *Corticium solani* showed symptoms similar to those described for the banded sclerotial disease of sugarcane. The disease was associated with that on *Cynodon dactylon* Pers. infected by *Corticium solani*. Ryker is of opinion that the fungus he was dealing with closely resembled the one found on rice and other plants in the East and variously described as *Hypochnus* (*Corticium*) *sasakii* and *Rhizoctonia* (*Corticium*) *solani*. A sheath rot and culm blight of rice occurring in many parts of the world (Dickson 1947) is variously attributed to *Corticium vagum* Berk. & Curt. *Hypochnus filamentosa* Pat., *Hypochnus solani* Prill. & Del., *Corticium solani*, and *Rhizoctonia solani*. Rogers (1943) considers all these to be synonymous with *Pellicularia filamentosa*.

Hemmi and Endo (1933) working on the sclerotial disease of rice found that there was no significant difference between the Japanese strain of *Hypochnus sasakii* and a Philippine collection of the same fungus but referred by M. A. Palo to *Rhizoctonia* (*Corticium*) *solani* group. Wei (1934) from his studies on the *Rhizoctonia* sheath blight of rice came to the conclusion that the fungus he was dealing with was identical with *Corticium sasakii* as well as with the Phillippine fungus determined by Palo as *Rhizoctonia* (*Corticium*) *solani*. He states that his fungus was a strain of *Hypochnus solani*. Matsumoto (1934) however is of opinion that *Corticium* (*Hypochnus*) *sasakii* and *Corticium solani* are two distinct fungi. *Corticium solani* has been known to occur on several other host plants and is also responsible for the " black scurf" of potato.

It will be noted that the fungus has a very wide host range and that it has been recorded from different parts of the world. Probably the existence of several strains with differences in colour and size of hyphæ and sclerotia and the occasional development of the basidial stage may have given rise to the several names attributed to it.

INFECTION EXPERIMENTS

The fungus was readily brought into culture by the transference of small bits of mycelial growth to oat agar plates. These were further purified by cultivation on water agar and isolation of single hyphal tips. The pure cultures were used for inoculation purposes. The experimental plants were grown in pots and kept inside glass cages or were covered over by bell jars to provide a humid environment. The results of infection experiments are described under each host.

Maranta arundinacea.—The inocula were placed on both the surfaces of young and old leaves. In all cases small water-soaked depressed spots developed within 48 hours. From these areas long aerial hyphæ were found to extend both above and below the infected region. The original spots extended and new irregular spots developed further on and in the course of six to eight days the entire leaf was affected. The spots turned dirty green to yellowish brown. If at this period the plants were moved into drier environment the progress of the lesions was arrested and the spots assumed a whitish colour. The characteristic formation of irregular bands stretching across the leaf was evident in some of the leaves. Under favourable conditions, rapid spread of infection took place. Besides the aerial hyphæ minute buff to tan sclerotioid bodies were seen on the under surface of the leaves. Sometimes the growth was lighter coloured and powdery. Infection did not spread to the lower portions of the shoots or the rhizomes. Infected leaves ultimately dried up.

Amomum sp.—Inoculations were made on one leaf of each plant. Lesions developed in 48 hours. In 12 days all the leaves (six) in each plant were involved. Characteristic banding was evident. Infection extended down the sheaths up to half

way down the shoots. Small dark buff sclerotioid bodies connected by long aerial hyphæ were formed on the lower surface of the leaves.

Canna indica L.—Evidence of infection was apparent in 24 hours as brown spots and in a week the entire leaf rotted. Infection did not extend down the sheath. In the initial stages, *i. e.*, after 24-48 hours, the hyphæ spread out on the leaf far in advance of the lesions as silvery threads.

Oryza sativa L.—Within 24 hours of inoculation the leaves of seedlings (12 days old) turned yellowish. In a week the sheath and leaves had rotted and turned brown and the seedlings had succumbed.

Zea mays L.—Young plants were used. Spots developed in 48 hours of inoculation and the leaves and the seedlings completely rotted in a week.

Setaria italica Beauv.—Within 24 hours spots began to develop on inoculated leaves. In one week all the plants were killed, the leaves being completely rotted.

Pennisetum typhoides. Stapf. & Hubb.—Plants one month old rapidly succumbed. On some of the leaves the characteristic banding with greyish green patches alternating with light green ones was seen. Others turned brown completely and rotted in a week. Dark brown speck like sclerotia were seen on the spots.

Saccharum officinarum L.—Young shoots were inoculated. Infection was apparent in 24 hours. The characteristic formation of irregular bands was clearly visible. The spindle rotted completely in 10 days.

Arachis hypogea Willd.—Infection was evident on the leaves in 48 hours. The leaves rotted and dropped down. The plants died in a week.

Cajanus cajan (Linn.) Millsp.—In 24 hours the leaflets had become discoloured and in four days they had rotted and were detached from the petiole. But these were held suspended by the cob-web like mycelium which had extended from the leaflets to the main stalk and stem.

Solanum melongena L.—Inoculations were carried out on both sides of the leaves. Dark water-soaked spots formed in 48 hours. Young terminal buds completely rotted in the same period. Defoliation of the infected leaves resulted in four days. Infection did not extend to the stem.

Lycopersicum esculentum Mill.—Spots developed on the leaves on the third day, but there was not the same rapid spread and enlargement of the spots as on *Solanum melongena.*

Citrus sp.—Brown spots were formed in 48 hours after inoculation. These enlarged quickly and the leaves dropped down when about three fourths of the leaf was overrun. Areolate markings were not observed; *Crinum asiaticum L., Caralluma sp. and Curcuma longa* L. were not infected by the fungus.

Suitable controls were kept in all the experiments and these remained healthy throughout. The inoculations were carried out on healthy unwounded leaves. It was observed that infection was very rapid in rainy seasons and when the plants were covered by bell jars. Successful infection results were obtained in all months of the year. The deciding factor for successful infection was the humidity of the environment. The results further showed that the fungus could readily infect a variety of plants belonging to several families and that it did not exhibit any specialisation of parasitism.

On the infected leaves the hyphæ extended both externally and internally. Sections of the affected leaves exhibited penetration by the hyphæ through the stomata into the mesophyll. The guard cells and the mesophyll cells in the vicinity of the hyphæ had lost their green colour, turned brown and collapsed. Meanwhile the hyphæ on the surface of the leaf had spread much farther and quicker. Fresh penetration into the leaf took place further ahead. This discontinuous penetration into the tissues by the quick growing aerial hyphæ was probably responsible for the formation of the banded appearance on some of the hosts.

CULTURAL STUDIES

The fungus grew readily on oat agar producing a thin stringy growth which covered up the surface of the medium in a petri dish of 10cm. diameter in 3 to 4 days. On oat agar slants also the surface was overgrown in three days, and the space between the wall of the tube and the surface of the slant was filled with loose hyphæ. In a week small hemispherical white cushiony sclerotioid bodies had formed along the edge of the growth in the dishes or the top and bottom of the slant. In addition big chocolate coloured sclerotia developed sparingly, six or seven per tube or ten to fifteen in a petri dish. Some of these had a diameter of 5—7mm. The surface of the sclerotia was velvetty.

On carrot agar the growth was very thin and only a few sclerotia developed. On an average eight sclerotia developed per petri dish in nine days. On onion-peptone-agar also the mycelial growth was thin and scanty. The sclerotial formation was similar to that on carrot agar.

The best growth was obtained on potato dextrose agar. Comparatively numerous sclerotia developed on this medium. Over 66 sclerotia were counted per dish in the course of 9 days. The perfect stage of the fungus did not develop on any of the agar media.

MORPHOLOGY OF THE FUNGUS

The mycelium is made up of branched hyphæ measuring 7-10μ in thickness. The branches develop usually at right angles but others making lesser angles with the parent hyphæ are also present. There is a distinct constriction at the point of origin of the branch. A partition wall usually develops 6—10μ beyond the constricted portion. Sometimes the septum forms at the constriction itself. Old hyphæ are empty and easily collapse. But occasionally a few hyphæ with thickened walls are also seen. Some of the hyphæ are completely filled with protoplasm and appear as glistening, solid septate segments under the microscope. These easily separate on teasing and later germinate, producing germ tubes which are at first much finer (less than 4—5μ in thickness) than the other hyphæ. Clamp connections are not observed in the hyphæ from cultures or in the hyphæ on host plants.

FIG. Basidial formation and basidiospores (x 250)

The sclerotia are hemispherical, flattened or irregular, and of varying sizes. They are isolated or in clumps. On the surface of the infected leaf these are usually small and buff to tan coloured. On agar media bigger sclerotia 5—7mm. in diameter are produced. These are chocolate coloured and have a velvetty surface, with one or two drops of fluid on the surface. When they become old round pits of varying depths are seen below the drops. The texture of the sclerotium as revealed in section is more or less uniform and made up of short irregular hyphal cells brown in colour mixed with darker elongated hyphæ. There is no differentiation into distinct cortical and medullary tissues as is common in many sclerotia of fungi.

Basidia are formed only on the surface of the leaves of diseased plants especially in the healthy unaffected portions and usually on the lower surface. The hymenial growth is whitish and consists of clusters distributed on a loose weft of hyphæ. Each cluster is made up of short branches of three or more series, the basidia developing in the ultimate series. From the base of the basidia fresh branches may arise. Thus a mixture of basidia and stout rounded branches can be seen. Each basidium is club shaped and from the apex four stout long sterigmata are formed. A hyaline elliptic or oblong one celled basidiospore is developed from each sterigma. The basidiospores have short papilla-like knobs (apiculate) and measure $7.5 \times 4.7\mu$ (7.2—10.8×3.6—7.2μ). The perfect stage usually develops in the month of August under field conditions.

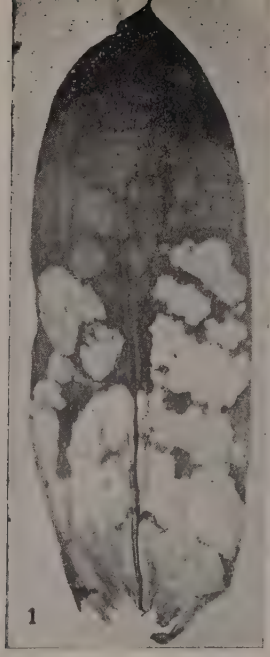
The morphological characters denote that the fungus is *Pellicularia filamentosa*. Rogers (1943) in his monograph on the genus *Pellicularia* has included under *Pellicularia filamentosa* a number of fungi previously described under the genera *Corticium*, *Hypochnus* and *Botryobasidium*. *Hypochnus filamentosa* Pat., *Hypochnus solani* Prill. and Del., *Corticium vagum* sensu Burt., *Corticium vagum* var. *solani* Burt., *Corticium vagum* sub sp. *solani* (Prill & Del.) Donk., *Corticium solani* (Prill. & Del.) Bourd. & Galz., *Corticium areolatum* Stahel, *Corticium microsclerotia* Weber, *Botryobasidium solani* (Prill & Del.) Donk and *Oidium citri* Bondar have all been made synonyms of *Pellicularia filamentosa*. Butler (1918) who has recorded the occurrence of the disease of *Maranta arundinacea* attributed the disease to *mycelia sterila* and stated that it may be *Corticium sasakii* (*Hypochnus sasakii*). It is quite probable that *Pellicularia filamentosa* was the pathogen in that case also. The natural occurrence of the perfect stage has enabled the identification of the pathogen in the present instance.

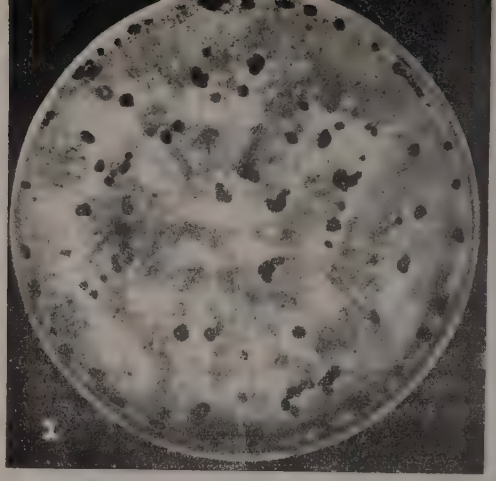
CONTROL MEASURES

Black rot of coffee caused by an allied species of fungus viz., *Pellicularia koleroga* Cke is prevalent in some of the coffee estates in South India. Venkatarayan (1947) reports that the best method of prevention of the disease is to spray the plants with one per cent Bordeaux mixture just before the south west monsoon. Both the surfaces of the leaves are to be covered and it may be necessary to repeat the spraying after the monsoon in September.

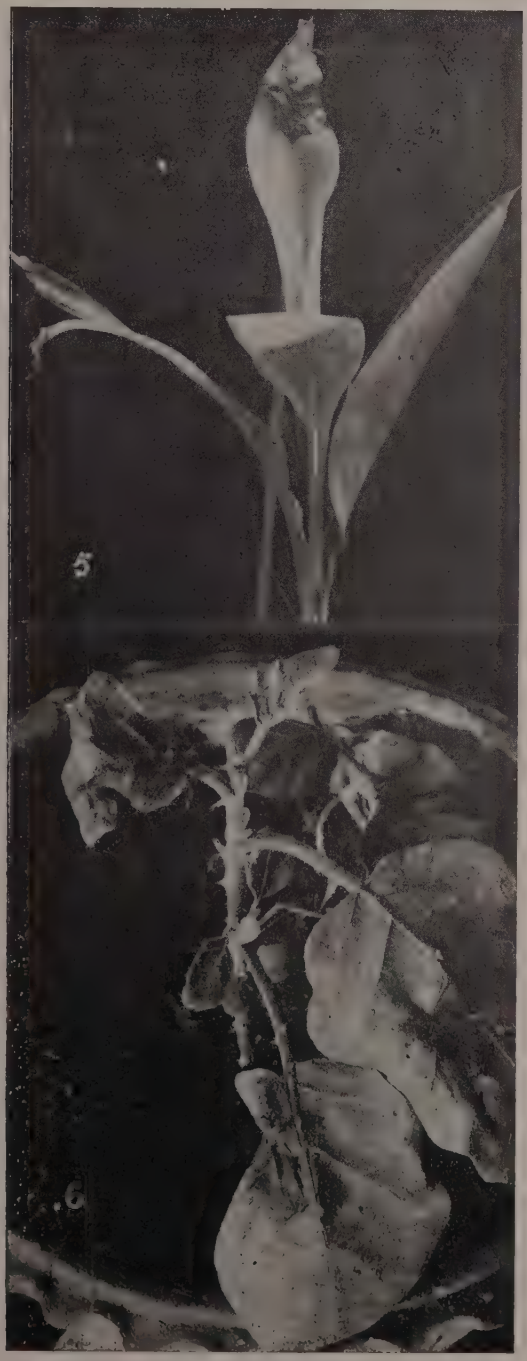
A similar treatment was carried out to control the disease under study. Four gardens were selected and preventive spraying with one per cent Bordeaux mixture was given in the month of July. In all the four gardens satisfactory control of the disease was obtained.* However, preventive spraying has to be carried out every year before the onset of the monsoon if the damage caused by the fungus is to be kept in check. Special care has to be bestowed to obtain complete coverage of both the surfaces of the leaves. The wide host range of the fungus enables it to survive on other hosts besides arrowroot. Consequently there is necessity for the adoption of the preventive measures every year.

* We are thankful to Mr. P. K. Nambiar, Agricultural Demonstrator, Cannanore, for carrying out the sprayings and furnishing the results.





1. Leaf of *Maranta arundinacca* showing symptoms of disease
2. Sclerotial formation on potato-dextrose agar
3. Symptoms of infection produced on inoculation on (3) citrus leaf, and (4) sugar cane leaf



5. Symptoms of infection on canna, and 6. on *Solanum melongena*

SUMMARY

A banded leaf blight of *Maranta arundinacea* is prevalent in North Malabar during the South-west monsoon. It is caused by *Pellicularia filamentosa*. The pathogen was brought into pure culture. Inoculation experiments showed that it had a wide host range. Timely spraying with Bordeaux mixture controlled the blight.

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BACTERIAL LEAF-SPOT OF DESMODIUM GANGETICUM DC.

By M. K. Patel and L. Moniz

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A bacterial leaf-spot of *Desmodium gangeticum* DC., was observed for the first time in 1943 at Bassein in Thana District. The host is a common road-side annual. The disease was not observed at any other place in Bombay Province and it would seem that it is localised at Bassein.

Gardner and Kendrick (1923) first described a white bacterial pathogen on *Desmodium canescens* and named it *Pseudomonas vignæ*. Later studies by Clara (1934) showed that this was a synonym of *Pseudomonas syringæ* Van Hall. Uppal and Patel (unpublished data) described a yellow organism on *Desmodium diffusum* and named it *Xanthomonas desmodii*. Except for these reports, no bacterial pathogen has been reported on other species of *Desmodium*. Since the pathogen on *Desmodium gangeticum* appeared to be quite distinct from *Xanthomonas desmodii* in symptom expression and pathogenicity, a study of its morphological, and biochemical characters was undertaken with a view to establishing its identity.

SYMPTOMS

The disease first appears as light yellow, water soaked spots on the undersides of leaves. These spots number a few to many on each leaf ; coalescence also occurs. The spots are generally round and measure upto a millmeter in diameter. As the disease progresses, the lesions turn greyish brown with necrotic centres and light yellow margins. These symptoms differ from those caused by *Xanthomonas desmodii* on *Desmodium diffusum*, in which case the spots are angular, yellowish brown, and limited by veins. Further, the spots on *Desmodium gangeticum* do not show any bacterial exudation and are noticeable only when viewed from the top. Infected leaves are generally distorted but do not shed. The disease is generally restricted to the leaves and petioles only.

THE PATHOGEN

The organism is a short rod with rounded ends, single or in pairs, but never in chains. Involution forms are absent. The average dimensions of cells from a three week old culture on potato dextrose agar are 2 (1.5—2.5) x 0.9 (0.7—1.4) microns. The organism is motile with a single polar flagellum, Gram-negative, non-sporulating, non-acid fast, but capsules are present. It stains readily with gentian violet, carbol fuchsin and methylene blue.

CULTURAL CHARACTERS

Colonies on potato dextrose agar plates are circular, smooth, glistening, convex and with entire margins. Colour is empire yellow (Ridgway) but odour is absent. Internal markings (striations) do not reach the periphery. On potato dextrose agar slants growth is copious, raised, smooth, glistening, filiform and opalescent with a butyrous consistency. Odour is absent. On nutrient agar slants growth is fair, slightly flat, dull, filiform, opalescent and lemon yellow in colour. Odour is absent. Moderate clouding is produced in nutrient broth in four days with some sediment

but no floccules or pellicle. Colour of the medium remains unchanged. On potato cylinders growth is abundant, orange yellow, flowing and covers the entire cylinder in a week. Colour of the cylinder is turned dark brown.

BIOCHEMICAL REACTIONS

The biochemical reactions of the organism were studied according to standard methods (Manual of Methods, 1936).

Gelatin liquefaction begins after 48 hours incubation and progresses rapidly. Hydrogen sulphide is produced. Indol is not formed. Nitrites and ammonia are not formed in six days. In litmus milk, complete reduction occurs in a week but tyrosine crystals are not formed. Plain milk is cleared in four days. The organism is a strict aerobe. Optimum temperature for growth is 20—25°C. with the minimum and maximum around 5° and 35°C. respectively, and the thermal death point is around 51°C. No growth in Cohn's medium and Koser's uric acid medium, but good growth was obtained in Simmon's citrate medium. Fair growth occurs in Uschinsky's medium. Growth is retarded by three per cent sodium chloride in nutrient broth and completely inhibited by a concentration of four per cent. Acetyl-methyl-carbinol is not produced (V. P. negative). The organism is able to utilise asparagin as the sole source of carbon and nitrogen. Lypolysis does not occur but the organism attacks starch and casein.

The following carbohydrates are utilised : dextrose, sucrose, raffinose, mannitol, salicin, galactose, maltose, sylose, lactose, arabinose and levulose.

Optimum pH for growth is around 7 ; range is 3.9—9.

PATHOGENICITY

In repeated trials, the organism failed to produce any symptoms on the following species when inoculated with pure cultures ; *Desmodium chephalotes*., *D. diffusum*, *D. gyrans*, *D. strobilaceum*, *D. triflorum*, *Arachis hypogaea*. *Cajanus cajan*, *Cicer arietinum*, *Crotolaria juncea*, *Cyamopsis psoraloides*, *Dolichos lablab*, *Lathyrus sativus*, *Medicago sativa*, *Phaseolus aureus*, *P. lunatus*, *P. mungo*, *P. vulgaris*, *Pisum sativum*, *Pueraria thunbergiana*, *Soja max*, *Trigonella faenumgrecum*, *Vicia faba*, *Vigna catjang*, *V. sinensis*, *Gossypium herbaceum*, *Lycopersicum esculentum*, *Solanum tuberosum*, and *Triticum vulgare*. Plants of *Desmodium gangeticum* inoculated at the same time produced typical symptoms in a week.

TAXONOMY AND NOMENCLATURE

The morphological, cultural and biochemical characters of the pathogen show that it is quite distinct from *X. desmodii* in many respects. A comparison between the two pathogens is made in Table I. Further, the two pathogens are not able to cross inoculate each other's hosts. Wernham (1948) made cross inoculation studies with 17 members of the genus *Xanthomonas* on 16 taxonomically distinct hosts and found that pathogenicity was a very specific characteristic of these species. His data indicated that, within the genus *Xanthomonas*, pathogenicity is of primary consideration for species demarcation, in the light of our present knowledge of the group. The data presented in this paper would indicate that the organism on *D. gangeticum* is very host specific and it is, therefore, intended to name it as a new

TABLE I

Comparative data for distinguishing morphological, cultural and physiological characters of the organism attacking D. gangeticum *and of* X. desmodii

Characters.	Organism from *D. gangeticum.*	*X. desmodii* Uppal & Patel
Pathogenicity on—		
Desmodium gangeticum	+	—
Desmodium diffusum	—	+
Dolichos lablab ..	—	—
Phaseolus vulgaris ..	—	—
Vigna catjang ..	—	—
General effects on hosts	Water-soaked, round spots with light yellow margin and necrotic centre	Water-soaked, angular spots with light yellow margin and necrotic centre
Morphology	Distinct capsule	Non-capsulated
Cultural characters—		
(a) Potato-dextrose agar plates	Growth smooth, convex, glistening. Colonies circular with entire margins. Internal markings (striations) not coming upto the periphery. Colour empire yellow 0.2 to 1.2 cm. in diameter with light yellow margins	Growth smooth, round, viscid, wet, shining, pulvinate, amber colour with colourless thinner margin of 1 to 2 cm. in diameter
(b) Potato-dextrose agar slants	Growth copious, raised, smooth glistening, filiform. Consistency butyrous. Colour empire yellow	Growth abundant, filiform, opaque, consistency butyrous. Colour amber yellow and growth not flowing
(c) Nutrient agar slant	Growth fair, dull, flat, opalescent, colour lemon-chrome	Growth fair, dull, smooth, flat. Coloured pinard yellow, colour of the medium unchanged
(d) Reaction in milk		
Plain milk ..	Growth moderate in 4 days with no pellicle and no sediment	Peptonized in 12 days
Litmus milk ..	Reduction slow, complete in 8 days with reddish tinge	Reduction of litmus slow in 10 days

\+ Means positive infection

— Means no infection

TABLE I—*concld*

Characters.	Organism from *D. gangeticum*.	*X. desmodii* Uppal & Patel.
Utilization of carbohydrates	Galactose, maltose, raffinose, mannitol, salicin, dextrose, sucrose, xylose, lactose, arabinose and levulose are utilised with acid in the first two and alkalinity in the next three	Acid but no gas in lactose, galactose and maltose. Growth poor in levulose, arabinose and xylose
Growth in asparagin ..	Good growth with yellowish round colonies measuring 3/4 cm. in diameter	The organism failed to grow
Growth at different pH..	Growth inhibited at pH 3.9 and below, while fair to moderate growth in pH ranging from 3.9 to 9	Growth inhibited at pH 8.5 and above. Heavy growth at 6.8 and 7.3 pH, while fair to moderate growth in pH ranging from 3.2 to 7.3

species, namely, *Xanthomonas desmodii-gangeticii* sp. nov., Uppal, Patel and Moniz. A technical description of the organism follows :—

Xanthomonas desmodii-gangeticii Uppal, Patel and Moniz sp. nov.,

Short rods with rounded ends, single or in pairs but never in chains. Capsules present. Motile with a single polar flagellum. Gram-negative. No spores and non-acid-fast. Stains readily with gentian violet, carbol fuchsin and methylene blue. Colonies on potato dextrose agar are circular with entire margins, smooth, convex, glistening and butyrous. Odour is absent and colour of the medium remains unchanged. Colour of the colonies is empire yellow. Internal markings (striations) not coming upto the margin. Moderate cloudiness in nutrient broth with no floccules and pellicle. Colour remains unchanged and odour is absent. Optimum temperature for growth is 20°—25°C. minimum about 5°C. and maximum about 35C. Thermal death point is between 50° and 52°C. The organism liquefies gelatin and has a strong diastatic action on starch. Casein is digested. Nitrates are not reduced and indole and ammonia are not produced. Hydrogen sulphide produced. Asparagin utilised as a sole source of carbon and nitrogen. Litmus reduced in litmus milk and plain milk is cleared. Utilises dextrose, sucrose, raffinose, galactose, maltose, xylose, lactose, arabinose, levulose, mannitol and salicin. Growth good in Uschinsky's solution ; no growth in Cohn's medium and Koser's uric acid medium.

The ogranism is pathogenic to *Desmodium gangeticum* DC.

SUMMARY

A bacterial leaf-spot of *Desmodium gangeticum* was first recorded at Bassein in Thana district. A species of *Xanthomonas* was isolated from the lesions on leaves and proved pathogenic on reinoculation.

The symptoms of the disease and the morphological, cultural and biochemical characters of the pathogen are described.

In inoculation trials in the green-house, the organism was pathogenic to *D. gangeticum* only and failed to produce any symptoms on any of the other species inoculated.

The organism is quite distinct from *X. desmodii* on *D. diffusum* and is therefore named as a new species namely *Xanthomonas desmodii-gangeticii* Uppal, Patel and Moniz, sp. nov.

A technical description of the organism is given.

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Indian Phytopathology 1-2:142,1948-1949.

STUDIES ON LENTIL RUST, UROMYCES FABAE (PERS.) DE BARY IN UP

By Raghubir Prasada and Upendra Nath Verma

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DURING 1945-46 and 1946-47 there was a severe out-break of rust, *Uro fabæ*, on lentil (*Lens esculenta* L.) at Delhi. The rust is common on minous crops in India and abroad. In India it is known to attack ler pea, sweet pea and broad bean as reported by Butler (1918) in the Indo- plain.

The life history of this rust had not been studied in India and little infor was available on the factors which caused the annual recurrence of this disea experiments reported in this paper were undertaken in order to elucidate these factors. Normally, lentil is sown in October—November and harv April. Pycnia and æcia of the rust, which is autoecious, are observed on t and stems in February. Within a few days of their appearance the disease rapidly but only æcia are found on the infected parts. Most of the infect do not show any pycnia, indicating the occurrence of secondary æcia. In the spread of the rust takes place by means of æciospores, the uredia appear late in the season, quickly followed by telia. The teliospores are formed in sorus as the urediospores. In cases of severe attack, even the legumes are to be infected. After the harvest there is, as a rule, no trace of either the host rust left in the field.

THE TELIAL STAGE

The uredial stage is followed by the formation of teliospores in the same sor and on the same mycelium. In nature, the teliospores are generally formed when the host reaches maturity and environmental conditions are unfavourable for the prop gation of the rust in the uredial stage. The teliospores are the resting spores in the but they may or may not require a dormancy period, depending on the rust and environmental conditions. With regard to the teliospores of *Uromyces fabæ* no formation is available regarding the resting stage, viability under different envir mental conditions and the precise temperatures that would permit their germi tion. Since this information is necessary to determine the role of teliospores in annual rust outbreaks, samples were collected from the field in April and stored sealed glass tubes at 3—5° C. 8—10° C. 14° C. and 17—18° C. A part of the collection was exposed to natural conditions and the rest kept indoors.

Soon after collection in April, infected leaves and bits of stem were soaked water overnight and teliospores were exposed to temperature of 7—8° C. 12—14° C. 17—18°C. 20—22°C. 29°C. and 30—35°C. for testing their germination. Within week about 25 percent germination was observed in the material exposed to tempe tures varying from 12° to 22°C. Such tests were conducted every month during the year with similar results, except that better germination, in a shorter period 2—3 days, was observed at 17—18° C. and the percentage of germination increased with the approach of winter. Tests made during January—February yielded better and more consistent results than those made earlier because environmental conditions were ideal for their germination at that time. Alternate wetting and drying hastened the process.

(Page 142 also appears facing p 127 !)

These observations show that teliospores of *Uromyces fabæ* do not require a resting period and that at a suitable temperature they can germinate soon after formation. In nature, however, this temperature and maximum humidity are available only during the succeeding winter and, for that reason, the teliospores retain their viability for long periods.

It is well known that the viability of teliospores is affected by exposure to high temperatures after formation. The loss of viability of teliospores of stem rust of wheat during summer was recorded by Stakman, Kirby and Thiel (1921), Lambert (1929) and Stakman (1934) in the Southern United States, and by Mehta (1940) in the plains of India. According to Prasada (1940, 1947) the teliospores of *Melampsora lini* lose their viability during summer that follows the harvest in the plains of India.

The effect of environmental conditions on the viability of teliospores of *Uromyces fabæ* was determined by exposing the material to natural conditions in the open after collection from the crop in April and storing part of it in the room and at lower temperatures (3—18°C.) in refrigerators. It was found that the teliospores stored at lower temperatures gave the best germination and retained their viability for nearly two years. Material exposed to natural conditions and kept in the room remained viable till February—March, *i.e.*, for nearly 10—11 months, and successfully infected young healthy plants of lentil. These results show that, unlike wheat stem rust and linseed rust, the teliospores of this rust withstand the summer heat in the plains of India.

For inoculations with teliospores, the technique employed was essentially the same as described by Newton and Johnson (1932). Inoculations made during November and December did not succeed, most probably due to unsuitable temperature; because teliospores kept for germination near the inoculated plants did not germinate whereas those kept at 12°—18°C. gave very good germination. Positive results were, however, obtained during January and February and pycnia appeared in 10—14 days and æcia in 15—20 days. Both young and old leaves of different ages of plants got infected and produced æcia in abundance. Ten to twelve days after the date of appearance of æcia it was observed that fresh æcia, not preceded by pycnia, were formed on plants in the neighbourhood of the initial infection. Inoculations made up to the end of February resulted in the formation of pycnia and æcia, whereas those made later in the season were not successful, apparently because temperatures above 22°C. are unfavourable for the germination of teliospores and infection of plants.

In order to see if infected pieces of host tissue bearing the teliospores could cause rust outbreak in the succeeding year when carried with the seed, experiments were conducted under controlled conditions. It was observed that pycnia and æcia appeared on all the plants where the seed had been sown mixed with infected debris, whereas plants raised from seed previously treated with Agrosan, to serve as control, remained healthy. This shows that the teliospores which are generally carried with the seed may cause fresh infection of the crop in the following year.

THE ÆCIAL STAGE

Inoculations made on healthy plants with æciospores resulted in the formation of secondary æcia in 8—10 days which were distinguishable from primary æcia by

the absence of pycnia. Inoculations were continued up to the fourth generation with a view to determine the stage at which uredia appear. It was observed that only æcia appeared in all the inoculations that were made during February but as the weather warmed up in March uredia began to be formed, irrespective of the age of plants. This gave an indication that the prevailing temperature governed the formation of æcia and uredia. Experiments carried out with detached leaf cultures (Clinton and McCormick, 1924), where 2—3 week old plants were inoculated with primary æciospores and incubated at different temperatures, show that secondary æcia appeared on shoots kept at 17—19° and 20—22°C. and uredia at 25°C. while no infection took place at 30°C. These results and the observation of uredia in the field, as the weather gets warm, show that the formation of uredia or secondary æcia in this rust is governed by the prevailing temperature and that the spread from plant to plant takes place mostly by means of æciospores, the urediospores playing a minor part in dissemination.

Inoculations made with æciospores from *Lens esculenta* infected *Pisum sativum*, *P. arvense* and *Vicia faba*, and æciospores from *Pisum sativum* infected *Lens esculenta*. Nearly 40 to 75 per cent germination of æciospores was obtained between 7° and 22° C. At 25° and 30°C. the spores did not germinate which probably explains the failure of infection during the latter half of March when the room temperature was 26°—29°C.

The longevity of æciospores at different temperatures was determined by keeping them at temperatures varying from 3°C to 30°C. They were found to remain viable at 3—8°C. 10—12°C. 17—18°C. 25°C. and 30°C. for 8, 6, 4, 3 and 2 weeks, respectively. Inoculations made with all the samples after six months gave negative results showing thereby that æciospores could not retain their viability from one crop season to the next.

THE UREDIAL STAGE

All the green parts including pods, leaves and stems are attacked by uredia. The sori grow on both sides of the leaves and the stems. They are small, scattered, roundish on the leaves and oblong on the stem, sub-epidermal and pale brown in colour. The epidermis in due course is ruptured and the urediospores lie exposed on the flattened bed of hyphæ. The spores are round to ovate, light brown, spiny with 3 or 4 germ pores and measure 20—30 x 18—26μ.

According to Hiratsuka (1934) the optimum temperature for the germination of urediospores of *Uromyces fabæ* f. sp. *Vici fabæ* is 16°—22·5°C. and the germination commences within 50 minutes at 20°—22°C. During the course of these studies 30—40 per cent germination was obtained at 7—8°C. 70—80 per cent at 17—18°C. 20—30 per cent at 21°—22°C. 10—15 per cent at 25°C.while there was no germination at 28—29°C. The optimum temperature was found to be 17—18°C. at which the spores started germinating in two hours.

As already stated, the uredia appear in March and by the end of the month the temperature in Delhi becomes unfavourable for maintaining rust cultures. Inoculations made on 5th, 6th and 12th March, when the temperature varied from 20° to 26°C. produced infection in 11—13 days whereas inoculations made on 26th and 29th March at 26—33°C. gave negative results.

The longevity of urediospores was determined during these studies. Infected leaves and stems were collected from the field in the middle of March, air-dried overnight in a room, and kept in paper envelopes and sealed glass tubes at different temperatures. Before storage the spores gave over 80 per cent germination. Viability tests were made every week at 17—18°C. The results show that the urediospores remain viable for 16—17 weeks when stored at 3—8°C. five weeks at 17—18°C. three weeks at 28—29°C. and two weeks at 36—37°C. A temperature of 7—8°C. was found to be most suitable for storing the urediospores. At the end of six months, however, no infection was obtained with any of these collections when the plants were inoculated in October—November.

DISCUSSION

When the present studies were started nothing was known about the life history of *Uromyces fabæ* infecting lentil, *Lens esculenta* L., in India and its annual recurrence. The results reported in this paper show that the uredia and æcia of this rust perish during the summer that follows the harvest but the telia withstand the heat with the result that the teliospores remain viable for nearly a year under natural conditions in the plains. The teliospores can germinate soon after formation at temperatures ranging from 12° to 22°C. Since, in nature, this range of temperature and high humidity are together available only during the following winter (January—February), the teliospores retain their viability until then. It has been demonstrated experimentally that teliospores mixed with the seed bring about infection of the plants in the following season, whereas plants raised from seed previously treated with a mercurial seed dressing remain healthy if protected from wind-borne infection. Therefore, burning the plant debris after harvest and treating the seed would probably control this rust considerably. Such a recommendation has also been made by Cass-Smith (1942) for flax rust in Australia.

It is noteworthy that æciospores give rise to æcia in successive generations and dissemination takes place chiefly by means of æciospores. Only the primary æcia are preceded by the formation of pycnia. The uredia appear rather late in the season and it has been shown that this is due to the influence of temperature. At 17°—19°C. and 20—22°C. secondary æcia are produced, whereas uredia are formed at 25°C. The uredia are of a very short duration and are quickly followed by telia.

SUMMARY

The outbreak of lentil rust, *Uromyces fabæ* was noticed on the crop in Delhi in February. The pycnia appeared first, followed by æcia. Secondary æcia were formed in great profusion and the rust was found to be disseminated through æciospores. Uredia appeared late in the season, quickly followed by telia.

The formation of æcia or uredia is governed by the prevailing temperature. Between 17° and 22°C. secondary æcia are produced, whereas uredia are formed at 25°C.

The teliospores do not require a period of rest and germinate between 12° and 22°C. They were found to retain their viability through the summer under natural conditions as well as in the room. At 3—18°C. they remain viable for two years.

Æcia were obtained by inoculating the plants with germinating teliospores during January—February. In artificial inoculations also æciospores produced æcia

in successive generations when the temperature was below 20—22°C. but at 25°C. uredia were formed. The æciospores germinate between 7° and 22°C and remain viable for two months at 3—8°C.

The uredia appear late in the season. The urediospores germinate between 7° and 22°C. with 17—18°C. as the optimum temperature. Under storage they remain viable for 16 weeks at 3—8°C.

It has been shown that æcia and uredia of this rust perish during the summer that follows the harvest but the telia withstand the heat, germinate during January when the weather is favourable and cause fresh rust outbreaks.

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PSEUDOMONAS MANGIFERÆ-INDICÆ, PATHOGENIC ON MANGO

By M. K. Patel, Y. S. Kulkarni and L. Moniz

(Accepted for publication November 10, 1948)

The mango is perhaps the fruit *par excellence* of India and some mangoes of this Province are unrivalled in any part of the world. In 1947, a bacterial disease was noticed on mango leaves on the Agricultural College Farm, Poona and in some gardens at Dharwar. Search through literature revealed that a similar bacterial disease has been described from South Africa by Doidge in 1915. Since the pathogen isolated at Poona differed in several morphological, cultural and biochemical characters from that described by Doidge (1915), studies were undertaken and are published in this paper. A short note by the authors (1948) was published earlier.

In India the disease has apparently existed from a long time. Correspondence with the Forest Botanist, Dehra Dun, revealed that on mango leaves in herbarium specimens collected by W. Johnston in Bihar in 1881, by Inayatullah Khan in Kheri and by J. Gamble at Dehra Dun in 1908, lesions similar to those on the Poona specimens have been observed.

Isolation of the organism

Young necrotic spots on mango leaves were selected and the pathogen was easily isolated from them by the usual poured plate method. Before attempting isolation, it was made sure that the spot gave out bacterial ooze under the microscope. This method was invariably successful in isolating the right type of organism.

Symptoms

The disease appears on the leaves as numerous small, angular, water-soaked areas (Fig. 1, A.), usually crowding towards the tip (Fig. 1, E.), and varying from 1 to 4mm. in diameter. These areas are, at first, light yellowish but later turn dark brown, with a clear halo around the necrotic spots. Several lesions are often found to coalesce, forming large necrotic spots. The surface of such spots is rough, and raised due to heavy bacterial exudation. In the case of very old leaves, the necrotic lesions become white and dry and get cracked. Many a time the marginal infection of leaves is very conspicuous resulting in deformities and cracking (Fig. 1, E.). Artificially infected fruits show at first water-soaked areas which later become black. Young heavily infected fruits are generally shed. In advanced stage of the disease, deep longitudinal cracks are formed on the fruit accompanied by heavy bacterial gummy exudations [(Fig. 1, D. (i).] Infection by wounding produoes diseased spots on tender stems, petioles and the stalks bearing the fruit. These spots turn into deep longitudinal scars accompanied by appreciable amount of bacterial gummy ooze. The incubation period in the case of leaves and fruits varies from 12 to 14 days.

Morphology

The bacterium is a short rod with rounded ends, single or in chains of 2 to 4 and has no involution forms. In culture on potato dextrose agar, varying in age from 1 to 3 weeks, the average dimensions are 0.89 (0.45 to 1.44)μ x 0.45 (0.36 to 0.54)μ. It is motile by one or two polar flagella, gram-negative, non-capsulated, not-acid-fast and a non-spore-former.

Cultural Characters

In nutrient agar plates, the growth after 24 hours is fair, flat, smooth, shining ; colonies round, with entire margins, creamy white, with no distinctive odour. After one week's growth, the border of the colonies becomes deeper white while the centre remains creamy white. On potato dextrose agar slants, the growth is copious, raised, smooth, glistening, filiform, opalescent, consistency butyrous, white to creamy-white with no distinctive odour. On potato dextrose agar (2 per cent.) plates, the colonies are smooth, pulvinate, glistening, circular with entire margins, (Fig. 1, B.) consistency butyrous, without distinctive odour, the medium being unchanged, with no internal markings or striations, white to creamy white (Ridgway) and measure 1.5 cm. after a week. On potato cylinders growth copious, flowing, raised, shining, covering the entire surface in 5 days; colour changed to light brown. In nutrient broth after 7 days, good cloudy growth with pellicle, without floccules but slight sediment is formed and colour is unchanged. In plain milk, no change takes place during first 4 days but after 7 days peptonisation starts. Litmus is completely reduced in 7 days with pellicle and gelatinous sediment at the bottom. On Endo's agar (Difco), the organism does not make any growth. In Starr's tyrosine media, the organism does not make any growth. The pathogen is a strict ærobe. The optimum temperature for growth is 20° to 25° C., growth ceasing at 5° and 35°C. The thermaldeath point is approximately 55°C.

Biochemical reactions

The organism liquefies gelatin and is able to digest starch and casein. The colour of the lypolytic medium is not changed, showing that it does not digest fats. Hydrogen sulphide produced ; nitrates not reduced ; indol and ammonia not produced ; M. R. and V. P. tests negative ; no growth in Cohn's medium and in asparagin but good growth in Uschinsky's medium after 7 days ; tolerant to 2 per cent sodium chloride ; Loefflers blood serum was completely liquefied in 5 days.

The organism grows well on several synthetic carbohydrate media, separately containing 1 per cent dextrose, lactose and sucrose, with production of acid but no gas, slight pellicle and sediment in 7 days. It makes slight growth in mannitol with slight production of acid while no growth takes place in salicin, l–arabinose, maltose, levulose, inulin and glycerol.

Pathogenicity

The organism is pathogenic to several varieties of grafted and country mangoes but not to maize, sorghum, cotton, tobacco, carrot, brinjal, tomato, bean, *Lathyrus sativus*, *Vinca* sp., *Sesamum orientale* and lemon. Cashew nut (*Anacardium occidentale*) is feebly susceptible.

Taxonomy and Nomenclature

The morphological, cultural and biochemical characters of the organism isolated from *Mangifera indica* show conclusively that it is quite different from *Bacillus mangiferæ* Doidge in several respects. A comparative statement giving the distinguishing characters of the two organisms is presented in Table I so as to bring out clearly their differences.

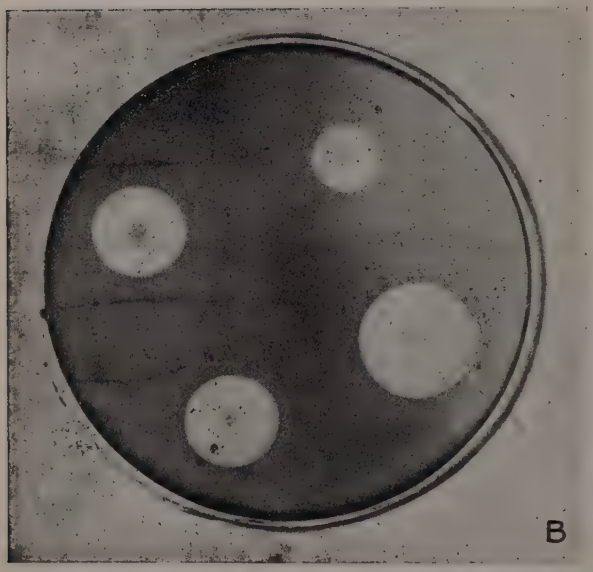
TABLE I

A comparative statement on morphological, cultural and biochemical characters of Ps. mangiferæ-indicæ *and* Bacillus mangiferæ.

Character	*Pseudomonas mangiferæ-indicæ*	*Bacillus mangiferæ*
Morphology	Gram-negative Non-capsulated	Gram-positive Capsulated
Cultural characters		
(i) Nutrient agar plates.	Growth slow, flat, smooth, shining; colonies round with entire margins, white to creamy white; border of the colony deeper in colour while centre remaining creamy white (Fig. 1, B)	Good growth in 24 hours; after 48 hours the colonies are 1 to 2 cm. in diameter, rather irregular in form with lobate margin. The colour of the colony is shining white for the first two days and after 5 days changes to maize or buff yellow (Fig. 1, C)
(ii) Potato cylinders	Growth copious, shining, raised, flowing and covering the entire surface in 5 days; cylinder changed to light brown	Growth only along the needle track, later spreading only at the lower part of the tube
(iii) Thermal death point	55° C.	60° C.
Biochemical reactions		
(i) Loeffler's blood serum	Liquefied	Not liquefied
(ii) Synthetic media		
(1) Uschinsky's	Good growth	No growth
(2) Cohn's ..	No growth	Slight growth
(iii) Starch	Hydrolysed	Not hydrolysed
(iv) Indol	Not produced	Produced
(v) Nitrate	Not reduced	Reduced
(vi) Hydrogen sulphide.	Produced	Not produced
(vii) Tolerance to NaCl	2 per cent	8 per cent
Fermentation of carbohydrates	No growth in levulose and glycerol	Slight production of acid in levulose and glycerol

According to recent classification advocated by Dowson (1942 and 1943), the phytopathogenic bacteria are split into four genera, viz., (1) *Bacterium*, (2) *Pseudomonas*, (3) *Xanthomonas* and (4) *Corynebacterium*. The organism under reference seems to fall into the genus *Pseudomonas* since it is whitish, gram-negative and a non-gas former in contrast to *Xanthomonas*, *Corynebacterium* and *Bacterium* which

Fig. 1, A-C. See legend for explanations



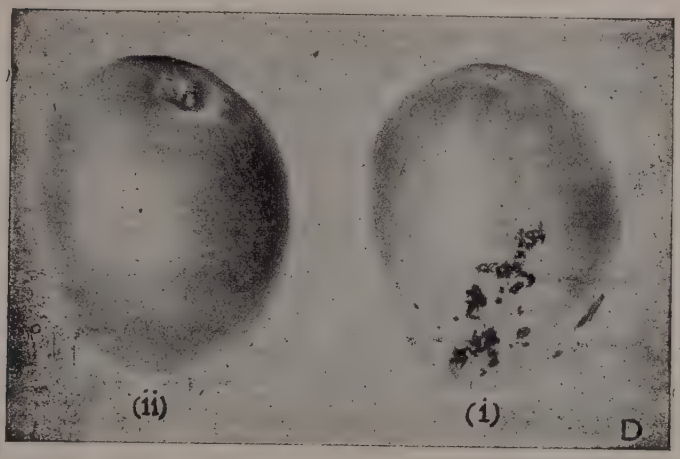


Fig. 1. D and E. See legend for explanations

are generally characterisd by yellow forms, gram-positive and gas formers respectively. Accordingly the pathogen has been named *Pseudomonas mangiferæ-indicæ* Patel, Moniz and Kulkarni (1948). The trinomial nomenclature had been adopted to show relationship to the common host and at the same time to distinguish it from *Bacillus mangiferæ* which although producing identical symptoms on *M. indica* differs in several morphological, cultural and biochemical characters (Table 1).

Pseudomonas mangiferæ-indicæ Patel, Moniz and Kulkarni sp. nov.

Short rods, single or in chains of 2 to 4, 0.45—1.44 x 0.36—0.54μ, no endospore, non-capsulated, gram-negative, motile.

On the potato dextrose agar, the colonies are circular, smooth, glistening, pulvinate, with entire margin, measuring 1 to 1.5cm. in diameter after 7 days ; white to creamy white ; no distinctive odour ; gelatin liquefied ; casein digested ; starch attacked ; hydrogen sulphide produced ; litmus reduced ; acid but no gas in dextrose, sucrose, lactose and mannitol ; M. R. and V. P. tests negative ; no growth in Cohn's and asparagin but good growth in Uschinsky's solution ; no production of nitrite, indol and ammonia ; Loeffler's blood serum liquefied in 5 days ; no growth in tyrosine medium ; non-lypolytic ; optimum temperature for growth 20° to 25°C. ; thermal death point about 55°C.

Pathogenic on *Mangifera indica* L., *Spondias mangiferæ* L and *Anacardium occidentale* L.

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Fig. 1. A. Mango leaf showing numerous, small, dark coloured, angular to irregular, water-soaked spots, characteristic of the disease. B. Round, smooth surface colonies of *Ps. mangiferæ-indicæ* on agar plates with characteristic creamy white centre and not flowing. C. surface colonies of *B. mangiferæ* in plate culture (photo from Doidge). D. Green fruits showing dark, rough, raised areas (gummy exudation) caused by *Ps. mangiferæ-indicæ*. Note the longitudinal cracking on the extreme right of the inoculated fruit (i) ; healthy fruit (ii). E. Mango leaves showing infection following the veins. Note cracking, distortion and crowding of spots towards the tip of the leaves.

SOME NEW HOSTS OF OIDIOPSIS TAURICA (LEV.) SALMON IN BOMBAY

By M. N. Kamat and M. K. Patel

(Accepted for publication Nov. 10, 1948)

Since the erection of the genus *Oidiopsis* by Scalia in 1902 and transfer of *Erysiphe taurica* Léveille by Salmon (1906) to that genus on account of its peculiar conidia and conidiophores and its endophytic habit, not much work has been reported regarding specialization in that fungus. Salmon (1906) established a variety and named it *Oidiopsis taurica* var. *lanuginosa* (Fuckel) Salmon on the basis of variations in the shape of conidia. Uppal, Kamat and Patel (1936) described a new variety, *Oidiopsis taurica* var. *macrospora* on *Dolichos lablab* because of the significantly larger conidia of their fungus than those of *Oidiopsis taurica* and the inability of that fungus to cross inoculate its other hosts. Since then the writers have collected a large number of hosts affected by a species of *Oidiopsis* and a detailed study of those collections is reported in this paper.

In recent years a tendency has developed among mycologists to create new species rather indiscriminately. New species must, as a rule, be based on characteristic and distinct morphological grounds. If this golden rule is not followed, a certain amount of confusion results. Thus Sawada (1934) raised the fungus attacking *Capsicum annuum* and *Papaver somniferum* to specific rank and gave them the names *Oidiopsis capsici* and *Oidiopsis papaver* respectively while Salmon (1906), Butler (1918) and Schweizer (1928) and many others have included the mildew attacking the former host under *Oidiopsis taurica*. Zeprometoff (1928, 1930) and Kvashnina (1928) created new varieties of *Oidiopsis taurica* to accomodate the endophytic mildews on *Carthamus tinctorius*, *Medicago sativa* and *Gossypium* sp., whereas *Zygophyllum fabago* and *Alhagi camelorum* are reported as hosts of *Oidiopsis taurica* by them as also Salmon (1906). The basis for the creation of these species and varieties by these authors is not, however, clearly understood.

Symptoms produced by this fungus on the new hosts on which the authors have collected this endophytic powdery mildew are similar to those produced on *Cyamopsis psoraloides* and other hosts described by Salmon (1906,) Butler (1918) and Uppal, Kamat and Patel (1936). For morphological studies 400 fresh conidia from these hosts were measured and the measurements compared with those of the conidia from *Cyamopsis psoraloides*. The results are tabulated in Table I.

TABLE I

Length of conidia of Oidiopsis *sp. on various hosts*

Classes in μ	Number of conidia in 400											
	Lycopersicum esculentum Mill.	*Martynia diandra* Glox.	*Crotalaria usaramoensis* Baker	*Euphorbia geniculata* Ort.	*Oxalis corniculata* L.	*Impatiens balsamina* L.	*Tagetes* sp.	*Phlox drummondii*	*Nasturtium* sp.	*Gynandropsis pentaphylla* DC.	*Cajanus cajan* (L.) Millsp	*Cyamopsis psoraloides* DC.
39 to 46.9	85	3	50	4	0	48	72	19	26	17	17	2
47 to 54.9	168	74	123	120	54	195	180	221	78	65	25	115
55 to 62.9	80	97	148	119	71	59	98	80	180	72	167	127
63 to 70.9	60	210	78	150	190	97	48	76	99	130	81	153
71 to 78.9	7	19	1	7	55	1	1	3	16	13	8	2
79 to 86.9	0	3	0	0	30	0	1	1	1	3	2	1
87 to 94.9	0	0	0	0	0	0	0	0	0	0	0	0
95 to 102.9	0	0	0	0	0	0	0	0	0	0	0	0
103 to 110.9	0	0	0	0	0	0	0	0	0	0	0	0
Total ..	400	400	400	400	400	400	400	400	400	300	300	400
	Width of conidia of Oidiopsis *sp. on various hosts.*											
4 to 9.9	10	5	15	0	0	4	0	0	0	0	0	0
10 to 15.9	247	384	350	333	135	324	122	24	194	77	151	123
16 to 21.9	143	11	35	67	265	72	272	375	205	223	134	277
22 to 27.9	0	0	0	0	0	0	6	1	1	0	15	0
28 to 33.9	0	0	0	0	0	0	0	0	0	0	0	0
Total ..	400	400	400	400	400	400	400	400	400	300	300	400

It will be seen from the results presented in Table I that the conidia from all the hosts fall within the range of 39 to 87μ which is also typical of *Oidiopsis taurica* on *Cyamopsis psoraloides*. As far as width is concerned, the fungus has a smaller range, viz. 4 to 28μ. The conidia of the fungus from *Oxalis corniculata* are slightly bigger than those from *Cyamopsis psoraloides* though they fall within the range of 32 to 82μ, typical of *Oidiopsis taurica* as stated by Salmon (1906).

Young vigorously growing plants of *Oxalis corniculata*, *Euphorbia geniculata*, *Tagetes* sp., *Cajanus cajan*, *Capsicum annuum* and *Cyamopsis psoraloides* were exposed to infection with fresh conidia of the fungus obtained from different hosts. Both inoculated and control plants were placed under bell-jars throughout the experiments to avoid aerial contamination from other sources. The results are given in the following table.

Table II

Cross inoculations tests with Oidiopsis *species*

Plants inoculated	Conidia taken from						
	Euphorbia	Oxalis	Impatiens	Tagetes	Cajanus	Cyamopsis	Capsicum
Impatiens balsamina ..	2/0	3/0	4/4	5/0	4/0	5/0	6/0
Tagates sp.	3/0	3/0	3/0	6/6	4/0	5/0	6/0
Cajanus cajan	2/0	3/0	3/0	5/0	3/3	4/0	6/0
Cyamopsis psoraloides ..	2/0	3/0	2/0	4/0	3/0	5/5	6/0
Capsicum annuum	2/0	3/0	4/0	4/0	3/0	5/0	10/9

* The numerator represents the number of plants inoculated and the denominator the number of plants infected.

It will be noted that the fungus is highly specialised in its parasitism as it infects only the host from which the inoculum was obtained and not other hosts.

The effect of temperature on the germination of the conidia of *Oidiopsis taurica* obtained from *Cyamopsis psoraloides* and *Euphorbia geniculata* was also studied. The results given in Table III clearly show that the optimum temperature for germination lies between 20° and 25°C. the range for germination being 14° to 35°C.

TABLE III

Effect of temperature on germination of conidia of Oidiopsis taurica *from* Cyamopsis psoraloides *and* Euphorbia geniculata

Temperature in °C.	Percent germination of conidia from		Remarks
	Cyamopsis psoraloiœes	*Euphorbia geniculata*	
5	0.0	0.0	
10	0·0	0.0	
14	trace	trace	
16	trace	2·5	
20	58·0	58·2	
25–26	47.3	51·5	
30	24·6	29·7	
35	2.4	1·9	
5 for 24 hrs. then 25–26	54.5	61.0	germ tubes vigorous

It will be further noted from the data recorded in Table III that when conidia are held at low temperature for 24 hours previous to their transfer to 24°—26°C, they give a higher per cent of germination and that the germ-tubes are also longer. It seems that exposure to low temperature stimulates germination as shown by Melhus (1911) for species of *Cystopus*.

GENERAL DISCUSSION AND CONCLUSIONS

Since the publications of Salmon (1906) and Butler (1918) reporting a wide range of hosts for *Oidiopsis taurica*, practically no work has been done on the distribution, host range and host specialization of this fungus. The present investigation adds 9 new hosts to the already existing long list and has shown conclusively that this fungus is highly specialized and as such is split up into several physiologic races.

Since the symptoms, shape and dimensions of the conidia of this fungus on the hosts mentioned herein are not significantly different, it may be referred to *Oidiopsis taurica* (Lèv.) Salmon.

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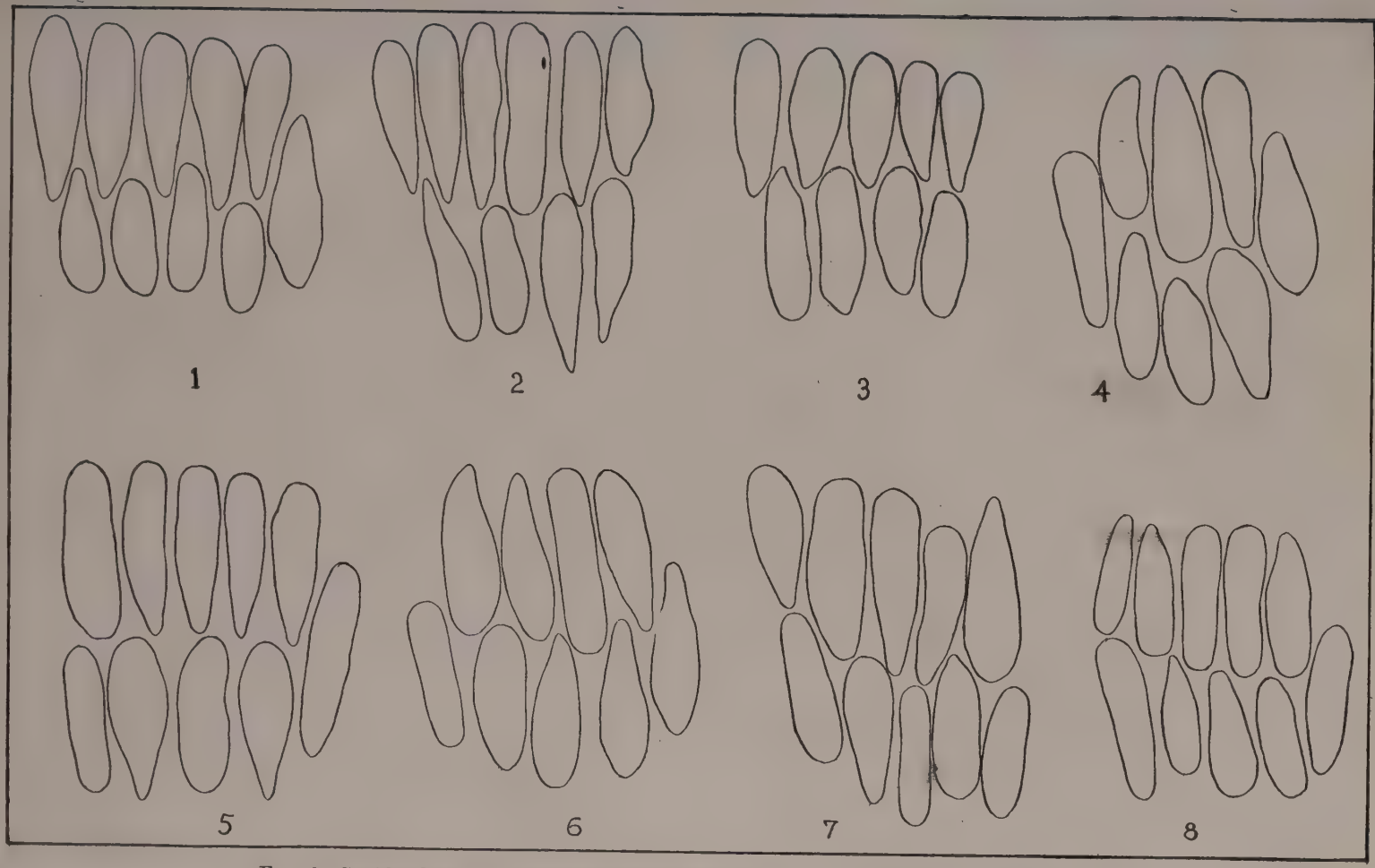

FIG. 1. Conidia from: 1. *Crotalaria usaramœnsis* 2. *Oxalis corniculata* 3. *Tagetes* sp. 4. *Cajanus cajan* 5. *Euphorbia geniculata* 6. *Gynandropsis pentaphylla* 7. *Martynia diandra* 8. *Phlox drummondii*

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BASIS OF PLANT QUARANTINES

By R. S. Mathur

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IT is now generally recognized that one important function of the Government of any civilized country is to protect and maintain the health of its agricultural crops against the ravages of insect pests and diseases. This is done by establishing a plant quarantine system and by sponsoring eradication programmes and various inspection services, depending upon the nature of pests and diseases and the extent of their spread. One important objective is to prevent the introduction of new insects and pathogens from foreign countries. In the United States, for example, the entry of citrus nursery stock from any part of the world is prohibited in order to exclude foreign diseases and parasites. If a pest or disease has already gained a limited foothold, the aim may be to eradicate it from the original center as was done in that country by promulgating plant quarantines for the complete eradication of the Mediterranean fruit fly and the citrus canker. Sometimes the aim is to stop the spread of a pest permanently, *e. g.*, peach mosaic, or it has to be delayed temporarily until better methods of control are available, as in the case of the Japanese beetle in the United States.

The purpose of this paper is to discuss the basis in logic for the plant quarantine laws.

CONTINUOUS AND DISCONTINUOUS SPREAD OF PESTS

From the plant quarantine point of view the term *pests* is used for all natural species inimical to human interests such as insects and their allies, fungi, bacteria, nematodes and viruses. With regard to their spread, the terms "continuous" and "discontinuous" spread were used by Butler (1917). It was *continuous spread* if the pests were disseminated from plant to plant or from field to field into contiguous areas by natural agencies such as wind, water, insects and animals, including man himself. Wind dissemination is common among rusts, mildews, smuts and many fungi causing leaf spots. Water, as wind-driven rain, spreads bacterial blight of cotton, red stripe of sugarcane and many other bacterial diseases. In floods, surface run-offs, streams and irrigation channels, water spreads many soil-borne organisms such as nematodes, damping off, and root rot fungi and some pathogenic bacteria. Insects as vectors of many virus, fungus and bacterial diseases are responsible for their spread to long distances. Birds carry the mucilaginous seed of dodders and mistletoes on their beaks and by wiping them on to the branches of new trees, spread these angiospermic parasites to wide areas. Man and his agricultural machines spread a number of diseases. Picking or cultivating beans while the plants are wet disseminates the bean blight bacteria. Pruning or transplanting operations and even smoking in tobacco fields, it is stated, spread the tobacco mosaic. Similar examples could be cited of the spread of insects in all their developmental stages through natural agencies. Insects possess wings and have the advantage of flight for their dissemination. It is extremely difficult if not impossible to control natural agencies responsible for the continuous spread of plant pests. In such cases, therefore, plant quarantines have a limited value. But where areas are separated by natural barriers, *e. g.*, high mountains, broad deserts, or extensive areas where the concerned host plant is absent, and the "discontinuous spread" of pests is due to the activities of man, in trade and

travel or in war and migration, plant quarantines play a very important part. Insects in all their developmental stages, *e. g.*, eggs, larvæ and imago, can be transported in various plant and animal commodities. Bacteria and the mycelium and the spores of fungi are effective sources of inoculum and can easily be carried on agricultural commodities, packing materials, soil attached to plants and even on the dress and baggage of tourists.

BIOLOGICAL CONSIDERATIONS

In nature, the host and its pests usually belong to the same place. Through centuries of exposure to pests, many species of susceptible plants are exterminated, leaving behind the more tolerant ones. Such surviving species tend to establish an equilibrium with their pests and environment, although this condition, called *biological equilibrium* by Orton and Beattie (1923), is generally loose. It is not a rigid mechanism to serve for the origin of resistant species. Plants under these conditions are at best "*klendusic*" or disease escaping and seem to tolerate better the injurious effects of pests in their natural habitat than plants not normally exposed to such conditions. If pests and pathogens are removed from their natural habitat this biological equilibrium is upset and the introduced pathogen is likely to become more destructive in a new country than it was in its original home. Smith *et al.* (1933), after an exhaustive study of the quarantine problems in California, arrived at the conclusion that, "with few exceptions such as grasshoppers, chinch bugs, potato beetle and some aphids, practically all the major pests and diseases of plants in the United States are those which have been accidentally introduced from foreign countries." It is therefore understandable that plant quarantine measures are largely applied to foreign pests. Research about the identity, life history, native hosts, extent and agencies of distribution and favorable and unfavorable climatic conditions for the development of the introduced pest is necessary. A knowledge of the susceptibility of its hosts, related species and varieties in the country of introduction and their economic value is also desirable. Investigations on all the biological aspects of the host and the parasite involve long-range problems and require years of study and research. This effort and expense is, however, fully compensated because plant quarantines promulgated on a sound biological basis are generally very efficaceous.

ECONOMIC CONSIDERATIONS

Most people are generally willing to see only the point of view which directly affects their well being. There are, therefore, many other points of view to the same question. Quarantines aim at the solution of long-range problems and are meant to benefit all society. The immediate gains or losses are distributed over long periods of time. Hence a consumer always benefits from plant quarantines which ensure abundance of production. As a citizen, an individual is affected by quarantines in a number of ways. He may be a grower owning infested crops or a consumer whose supply of agricultural goods is affected by pest damage. He may be a business man dealing in agricultural products ; he may be an exporter or an importer or a manufacturer of fungicides and insecticides. He may be a quarantine official, a county agent, an extension worker, a community leader, a writer, teacher, lawyer, witness, judge, or a juror. In all these capacities he may have to understand or explain the true significance of plant quarantines. In the true economy of a country the interests of individuals or groups should be complementary, and the economic consequences of a quarantine should be properly balanced. Otherwise incidental trade advantages and monopolies or commercial disadvantages may result by the enforcement of

quarantines in retaliation. Chester (1947) points out that, "A quarantine enacted primarily as a trade barrier for the purpose of protecting a local industry or as a retaliatory act of one state or country against another, disguised as a pest control measure, except for the protected minority, the gross economic loss may be even greater besides arousing prejudice against legitimate disease regulation." Public support and popularity are absolutely necessary for the successful functioning of a plant quarantine. This is possible only when the different economic factors are so compromised as to yield the maximum returns to the public in general. If a balance sheet of quarantines were examined, the debit side would represent :

(1) High unit cost of production due to the reduced yields caused by the pests or pathogens ;

(2) The cost of research, administration, publicity and enforcement of plant quarantines ;

(3) Losses of markets on account of retaliatory measures ;

(4) Losses due to the adjustments to new types of crops and agricultural methods ;

(5) Losses due to the eradication, condemnation and abandonment of crops.

On the credit side of plant quarantines may be considered :

(1) Freedom of crops from diseases and pests resulting in the abundance of production and lowering of prices without lowering profits of the producer ;

(2) Savings in labor and cost of direct control measures which may have to be applied in the absence of quarantine ;

(3) Trade advantages to growers outside the quarantine area.

It would be seen that the economic advantages arising out of plant quarantines far outweigh the investment in their enforcement. Smith *et al.* (1933) state that, "The policy of using police power to exclude dangerous plant pests and diseases is based on the premise that economically it is better to undergo considerable inconvenience and initial expense in an effort to exclude a pest or disease than to submit to the expense of controlling it for an indefinite period." In the United States plant quarantines are periodically reviewed and the economic gains or losses carefully assessed so that a quarantine which has served its useful purpose is promptly rescinded or modified under changed conditions. The economic justification of plant quarantine in that country has, therefore, been rarely questioned.

THE NECESSITY OF PLANT QUARANTINES

Some pests and diseases like the San José scale and the potato blight are so widespread that they can best be controlled by individual effort. Others require technical help and knowledge of the life cycle of the pest or parasite, *e. g.*, Mormon cricket, phony peach and apple scab. Still others like the white pine blister rust, Dutch elm disease, citrus canker, Japanese beetle, etc., where control, prevention or delay in spread from the original point of infestation involve community effort

and governmental help. The control of such pests and diaseases requires high technical skill or is ineffective unless applied on a large scale. It is also possible in such cases that the infection comes from an area far away from an individual's holding or that the interests of other areas are likely to suffer. Exclusion of a foreign pest or pathogen is an important basic principle. But in cases where the pest is already established its spread should be prevented or delayed. Where it has spread widely, its suppression, eradication or control may be necessary. In order that these objectives may be achieved, many restrictions have to be imposed on the entry or free movement of agricultural commodities and materials likely to serve as media for the transference of pests and pathogens. Such interference with the normal activities of man is brought about by *quarantines*. In other words, a quarantine is a legal restriction on the movement of commodities for the purpose of exclusion or the prevention or delay in the establishment of plant pests and diseases in areas where they are not known to occur. The enforcement of quarantines requires considerable expenditure of money and much interference in trade, travel and other normal activities of man. There should be, therefore, ample justification and an urgent need for them. The National Plant Board of the United States (1932) laid down the following four important prerequisites for the establishment of a quarantine :

(1) "The pest concerned must be of such nature as to offer actual or suspected threat to substantial interests ;

(2) "The proposed quarantine must represent a necessary or desirable measure for which no other substitute, involving less interference with normal activities, is available ;

(3) "The objective of the quarantine, either for preventing introduction or for limiting spread, must be reasonable of expectation ;

(4) "The economic gains expected must outweigh the cost of administration and the interference with normal activities."

PUBLIC SUPPORT AND WORKABILITY OF PLANT QUARANTINES

Plant quarantines have to be expressed in very clear language so that everybody can understand the duties involved in their successful working. The procedures should be simple and should cause the least interference with normal activities which will serve the purpose of quarantine. Before a quarantine law is passed, there should be a notice and a public hearing. The legal sanction, jurisdiction and police power entrusted to quarantine agents should be clearly specified. The government should also maintain a research and an information service for the stimulation and development of public understanding. The publication of bulletins, articles, press notes, etc., should be encouraged. Conferences, discussions, radio talks, seminars, exhibits and in fact any activity which may help a thorough understanding and concerted action of a community are very desirable. Intelligent leadership by government agents in these matters creates public sentiment, inspires support and leads to good planning and quicker results. According to McCubbin, (1944) "a plant quarantine is to be regarded as a *community undertaking* whereby the preponderance of public opinion supports its administrative representatives in imposing restrictions designed to prevent or limit the introduction or spread of injurious plant pests so as to promote the public welfare." The results arising out of quarantine enforcement should be periodically reviewed so that it could be modified or repealed as circumstances change.

QUARANTINE LAWS AND AGENTS

In the United States the federal regulatory measures authorized under the Plant Quarantine Act of 1912 are administered by the Bureau of Entomology and Plant Quarantine. The federal quarantine laws are of two categories :

(1) *Foreign quarantines* which prohibit or regulate the entry of imports from other countries, necessitating international co-operation on phytopathological and entomological problems ;

(2) *Domestic quarantines* which regulate inter-state shipments within the country.

Besides the federal quarantine, every state in the Union has its own laws administered by the state authorities prohibiting or regulating the movement of certain plants into or within the state. For certain types of materials inspection, fumigation or both are required. Re-inspection is done in many instances at the point of destination.

SUMMARY

The fight against crop pests and diseases by the use of regulatory measures is an important modern development in plant pathology.

Pests are disseminated by *continuous* and *discontinuous* spread. *Continuous* spread takes place by natural agencies over which we have little control. *Discontinuous* spread of pests is brought about by human activity which has to be restricted by passing plant quarantine laws in order to prevent the introduction of foreign pests. Other objectives of plant quarantines may be to prevent permanently or delay the spread of pests if they are already established in restricted areas.

The biological and economic considerations which form the basis of most quarantines have been discussed.

ACKNOWLEDGMENTS

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OCCURRENCE OF LATE BLIGHT OF POTATO IN THE PLAINS OF INDIA

By T. B. Lal

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LATE blight of potato had not been reported from the plains of India till 1899-1900, when it was for the first time observed in a few fields in the Hooghly district. The disease spread throughout the whole district in 1901—1902 but was not heard of from anywhere in the plains of India for about a decade. In 1912—1913 it was reported from Jorhat (Assam), in 1913 from Rangpur (Bengal) and Sabour and Bhagalpur (Bihar) and in 1916 from Karimganj (Sylhet). It was recorded at Pusa (Bihar) in February 1928. The latest record of the disease in the plains,until 1943, was made early in February 1933 at Bibiganj Bhatta, Patna, where it was noted in a small plot.

During the course of survey of the potato crop in the plains of Northern India during 1942—43, late blight was observed for the first time at Meerut and at Dehra Dun in March 1943. The varieties affected at Meerut and Dehra Dun were 'Gola' and 'Tumri' respectively. It is of interest to note that the disease was absent in the first crop (September-October to December-January) but it appeared in March, that is, in the second crop (January-February to April-May).

The crop had been examined in February but no trace of the disease had been observed. On March 5, 1943 about 1.4% of the plants at Meerut showed typical symptoms of late blight. In one plot in particular infection was upto 56%. At Dehra Dun late blight was noted on two plants only on March 13, 1943.

During the second inspection of the potato crop in these localities, in the first week of April 1943, incidence of late blight at Meerut and Dehra Dun was found to be 9.3% and 1·3% respectively. In one plot at Meerut, however, infection was as much as 29.5%.

At Meerut both late blight and early blight were found on the same leaf. Such combined infection varied from 1.2 to 1.6% during the two inspections.

It is now well established that the causal fungus (*Phytophthora infestans* (Mont.) de Bary) is unable to survive the heat of the summar months in the plains both in the soil and in the tubers. Origin of the appearance of the disease in the second crop at Meerut and Dehra Dun may be traced to the sowing of seed tubers from infected plants which had been imported, prior to sowing, in the months of December and January from the hills in the Dehra Dun district and Tehri State. Such tubers had apparently not been exposed to high summer temperatures of the plains before planting and the fungus therefore continued to remain in a viable state. The absence of the disease in the first crop at these two places is apparently due to the fact that the seed came from the second crop grown at Meerut and Dehra Dun in the preceding season. The period of exposure to high temperatures was thus adequate enough to kill the fungus, if it was at all present.

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MORPHOLOGY AND CYTOLOGY OF TILLETIA HOLCI ON HOLCUS MOLLIS L

By M. C. Das

(Accepted for publication December 20, 1948)

GERMINATION of chlamydospores of the following species of the genus *Tilletia* is known :—*Tilletia decipiens* (Pers.) Wint., (Brefeld—1895, Cunningham 1924), *Tilletia fœtida* (Wallroth) Liro, (Wolff-1874, Clinton 1902, Kiehnholz & Heald-1930, and other workers), *Tilletia fusca* Ell & Ev., and *Tilletia hordei* Koern. (McAlpine—1910), *Tilletia horrida* Takahashi (= *Neovossia horrida* (Tak.,) Padwick & Khan), (Teng-1931), but nuclear details are lacking. The most thoroughly studied species are however *Tilletia caries* (DC) Tul. and *Tilletia fœtida* in which the life history and nucleat details were extensively investigated by Dangeard (1892) Harper (1899), Lutman (1910), Parvicini (1917), Rawitscher (1914, 1922), Dastur (1921), Kharbush (1927), Boss (1927), Buller and Vanterpool (1933), Wang (1934) and others.

From the studies of the above investigators, the life cycle of the species of *Tilletia* can be represented as follows :—The mature chlamydospore germinates giving rise to a promycelium. The diploid nucleus of the spore undergoes divisions resulting in the production of 4 to 16 haploid nuclei. At the tip of the promycelium a cluster of primary sporidia is developed. The haploid nuclei migrate to the sporidia, conjugation of the sporidia follows in pairs, the nucleus of one of a fusing pair of sporidia passes to the other giving rise to a binucleate sporidium, which in turn produces a secondary sporidium, or gives rise to a dicaryotic hypha. The secondary sporidia are stated to be uni-or bi-nucleate.

Text Fig. 1

Spore formation in this genus was first studied by Fisher Von Waldheim (1869) in *Tilletia olida* (Riess.) Wint. Wang (1934) and others have described the process of chlamydospore development in *Tilletia caries* in the tissues of the host plant. In the genus *Tilletia* chlamydospores are formed singly at the end cells of the sporing hyphæ which usually contain two nuclei. These cells swell, enlarge and nuclear fusion takes place giving rise to young chlamydospores. In the process of maturation these spores acquire a double wall and the epispore is covered by reticulate thickening.

As there is no record of the germination of the chlamydospores of *Tilletia holci* (Westend). Schroet., and since little is known of its life cycle, the present investigation was undertaken with a view to working out morphological and cytological details of the species.

MATERIAL AND METHODS

Smutted panicles of *Holcus mollis* parasitised by *T. holci*, were collected from Linlithgow and other localities near Edinburgh. Mature chlamydospores were placed for germination in different solid and liquid media, as will be described under germination. When the desired stages in the germination of the spores were reached, the material was killed and fixed in Flemming's weak solution or Karpechenko's fixative. The material fixed in Flemming's was bleached with 10 per cent H_2O_2 for about 20 to 30 minutes, and stained with Flemming's triple stains, or with iron-alum-hæmatoxylin with slight modifications and variations.

To study the nuclear condition of the parasitic mycelium and the development of the chlamydospores in the host tissues, infected rhizome, stem, and panicles of *Holcus mollis* at various stages of parasitization, were fixed in Flemming's weak solution. Microtome sections from 6 to 12μ were cut, bleached with 10 per cent H_2O_2 for 8 to 12 hours before staining, and stained with Bismarck brown-gentian violet followed by Gram's iodine, or Flemming's triple stain, or with iron-alum-hæmatoxylin in the usual way. Flemming's triple stains and iron-alum-hæmatoxylin yielded the best results.

MORPHOLOGY

Sori are formed in the ovaries replacing the seeds and enclosed in the pericarp. They are partially concealed within the outer glumes. Almost all the spikelets of the panicle are attacked and destroyed (Text Fig. 1). Each spikelet very often consists of two hermaphrodite flowers, and both the ovaries are attacked and transformed into powdery brownish black spore masses. Sori are more or less elliptical, and about 1 to 2mm. long and 1mm. wide Upon rapture, they

Text Fig. 2

release the powdery, yellowish-brown to olive-brown, deeply reticulate spores. Spores are chiefly globose but occasionally they are subglobose ; the epispore is covered with a network of reticulations, surrounding almost polygonal depressions. The reticulations swell when the spores are soaked in water for some days. They measure 3—8μ in thickness. There is a good deal of variation as regards the size and colour of the spores (Text fig. 2). They measure 22—35μ in the outer diameter and 16—24μ in inner diameter. Generally the deep brown spores have a larger inner diameter but low reticulations (3 to 4μ) (Text fig. 2). Olive brown spores have smaller or slightly smaller inner diameter and high reticulations (4 to 8μ). Between the two extreme types, all kinds of variations may be found. Occasionally some sterile spores with smooth or reticulate walls may be seen. These sterile spores are mostly globose to somewhat angular, hyaline to pale smoky yellow in colour.

GERMINATION OF CHLAMYDOSPORES

Considerable difficulties were experienced in germinating the chlamydospores. Various kinds of solid and liquid media, including various concentrations of Aneurine (Vitamin B_1 'Roche') were tried. The results are shown in Table I.

TABLE I

Germination of chlamydospores of Tilletia holci

Media	Temp.	Results
1. Distilled water	18—20°C.	No germination up to 45 days
2. Spore extract (chlamydospores ground with fine sand and filtered through a porcelain filter)	,,	A few spores germinated after 40 days.
3. Yeast extract (Yeast obtained from Brewery, ground with fine sand and filtered through a porcelain filter)	,,	No germination up to45 days.
4. 0.3 per cent Calcium Nitrate solution	,,	No germination up to 45 days
5. 1.5‰.....KH_2PO_4 0.5‰.....$MgSO_4$ 1.5‰.....Asparagin 1.0%.....Glucose To this solution various concenrattions (500.0mmg. 100.0mmg., 25.0mmg., 5.0mmg., 1.0mmg., 0.05mmg., 0.25mmg., 0.1mmg, 0.05mmg., 0.025mmg.) of Aneurine were added	,,	Only a few spores germinated after 35 days at concentrations of 0.05 mmg. and 0.025 mmg.
6. Tap water	,,	No germination up to45 days.
7. Spores bleached with 10% bleaching powder solution for 3 to 5 minutes and placed in sterile tap water in hanging drops and kept in moist petri dishes and allowed to stay outside for 5 to 7 days at a temp. 1°C. to 3°C. and then removed to the room temperature (18-20°C.)		50 to 80 per cent germination obtained within 48 hours.
8. Spores subjected to the same treatment as in 7 and placed in 1% agar slants.	,,	50 to 70 per cent germination obtained within 48 hours.

Fresh spores, germinating spores, primary sporidia and secondary sporidia placed for germination and growth on the various solid media such as 2% agar, 2% malt extract agar, 2% potato dextrose, 2% glucose, soil extract agar, did not yield satisfactory results. On 1% agar slants, as noted above, spores germinated and produced primary and secondary sporidia and some hyphæ but there was no further growth.

CYTOLOGY

Like other smut spores, the chlamydospores of *Tilletia holci* contains a diploid nucleus (Pl. I fig. 1), which is usually situated at the centre of the spore. The cytoplasm contains a few large vacuoles, and some dark staining bodies distributed evenly in the cytoplasm. The first sign of germination can be observed when the episore, with its reticulations, cracks open and a thick tube begins to push out from within (Pl. I fig. 4). This tube continues to grow giving rise to a long or short promycelium. The promycelium at first remains thick, but as it grows it thins out a little and becomes more or less uniform. It varies in length but is more or less constant in width, about 6 to 7μ. Some promycelia produce primary sporidia when they are very short, while others grow to variable lengths. Some of them have been observed to grow as long as 700μ in length without showing any signs of forming sporidia. Often the promycelium branches (Pl. I fig. 13). Branching continues for some time until from one of the branches the sporidia develop. Usually one promycelium is produced from each spore, but rarely two, which grow for a short time, then the contents of one promycelium migrate to the other, leaving behind an empty one. A similar movement of protoplast from one branch of a promycelium to another branch of the same has been constantly observed when branching occurs. No partition wall, either in the promycelium or in the branches, has been observed dividing the living protoplast.

As the promycelium grows, its contents move forward towards the tip, leaving behind an empty space which is cut off by a concave-convex wall. At the time of formation of the wall, there appears a small vacuole at the basal part of the protoplasmic content of the promycelium, which soon begins to grow and is cut off by a wall. Ultimately the vacuole increases in size and the contents disappear and the whole vacuole becomes an empty cell. A similar mode of development of an empty cell at the basal part of the promycelium in *Tilletis caries* has been described by Buller and Vanterpool (1933). As the promycelium grows in length and as the protoplasmic contents keep on moving forward, the number of empty cells in the basal part goes on increasing.

Owing to the colour and thickness of the epispores it was extremely difficult to observe the nuclear condition inside the spore. It was not clearly understood why iron-alum-hæmatoxylin failed to give a satisfactory stain to the nuclei inside the spore, and in the promycelium. However, materials stained with gentian violet for 24 hours, after mordanting with Gram's iodin yielded satisfactory results. Especially the nuclei in the promycelia, sporidia and hyphæ stained most satisfactorily. With the onset of germination, the diploid nucleus of the spore undergoes certain changes, resulting in the formation of several round or oval shaped bodies (Pl. I figs. 2, 3). Along with the flow of the contents of the spore to the promycelium during germination, these bodies—sometimes 2, sometimes more (Pl. I figs. 4, 6)—come out to the promycelium. These bodies are none other than the nuclei, as can be recognised from their size, shape and further activities in undergoing divisions in the promycelium. This clearly and definitely indicates that the diploid nucleus divides once, twice or sometimes more inside the spore, before or during the development of the promycelium. Sometimes however, the diploid nucleus migrates to the promycelium where it divides (Pl. I fig. 7).

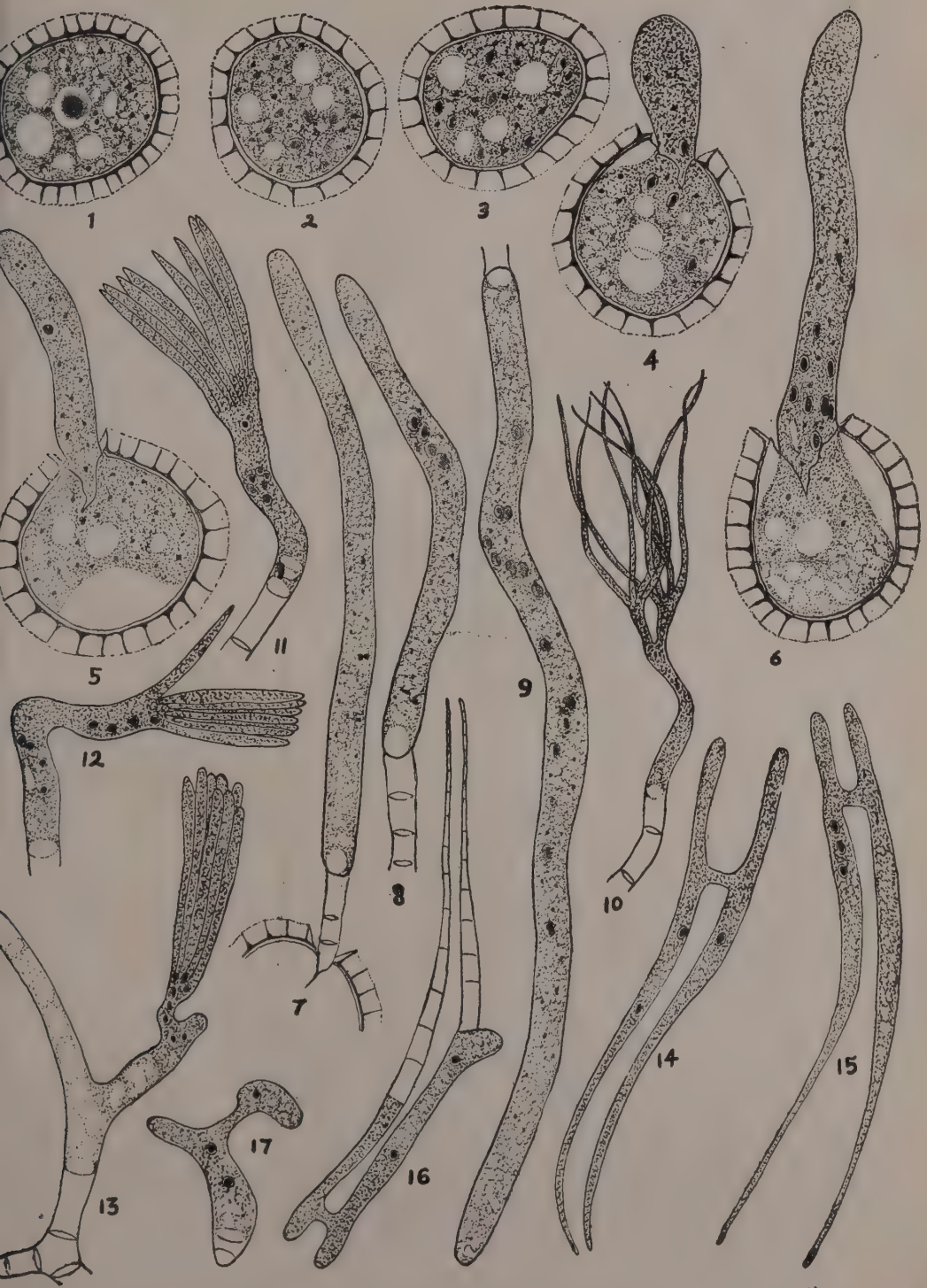
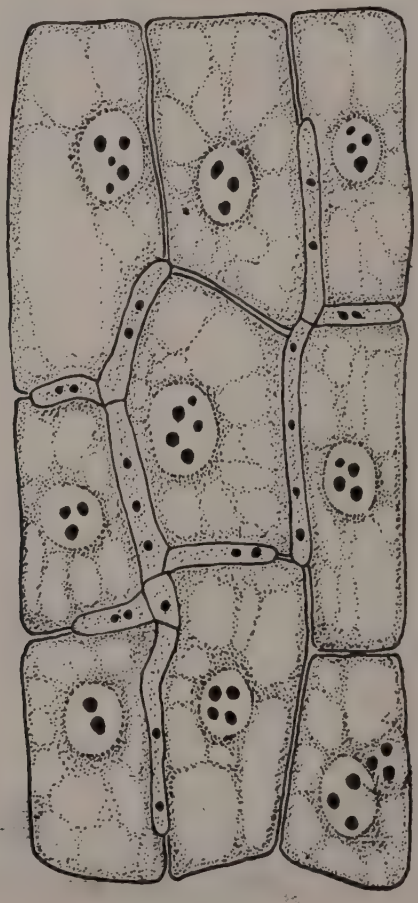


Plate I. Figs. 1 to 17. See legend for explanation

There is a good deal of controversy as to where the first division of the diploid nucleus of *Tilletia caries* takes place. Dangeard (1892), Dastur (1921) and Kharbush (1928) claim that the first division takes place in the promycelium, but Parvicini (1917) and Rawitscher (1914, 1922) hold that the first division of the diploid nucleus is completed inside the spore. Observations of Wang (1934) and Holton & Heald (1941) show that the first division of the diploid nucleus of *Tilletia caries* may take place either inside the spore or in the promycelium. Observations made on a great number of germinating spores of *Tilletia holci* agree with those of Wang (1934) and Holton and Heald (1941) in *Tilletia caries*. In *Tilletia holci* the first division may take place in the spore or in the promycelium.



Text. Fig. 3

When the diploid nucleus divides in the promycelium it is at first resolved into four chromosomes (Pl. I fig. 7). Unfortunately, the further behaviour of these chromosomes resulting in the production of two daughter nuclei could not be worked out in detail owing to the poor visibility of the chromosomes in the stained preparations. However, a few cases were noted where the daughter nuclei are in a state of division.

Certain preparations show that the chromosomes of a daughter nucleus separate into two groups, each group containing 2 chromosomes (Pl. I, fig. 5). This indicates that the haploid number of chromosomes in *Tilletia holci* is two. The two daughter nuclei soon divide successively or simultaneously giving rise to four nuclei (Pl. I, fig. 8), which in turn divide again giving rise to eight nuclei. Usually the sporidia are developed at this stage, but occasionally another division follows resulting in the production of sixteen nuclei for the mature promycelium.

The time and place of second and subsequent divisions of the diploid nucleus is variable like the first one. All other divisions may take place either in the spore or in the promycelium at some stage subsequent to germination. Whether some divisions are in the spore and others are in the promycelium depends upon the time when each nucleus passes into the promycelium. Moreover, the divisions of the nuclei of the same generation are not usually simultaneous, for one can see promycelia with an odd number of nuclei. A similar mode of second and subsequent divisions of the diploid nucleus in *Tilletia caries* has been described by Wang (1934) and Holton and Heald (1941).

Paravicini (1917) thought that the bifurcation of the tip of the promycelium is connected with the second and subsequent divisions of the diploid nucleus in *Tilletia caries*, but Dangeard (1892), Rawitscher (1914, 1922), Dastur (1921) and Wang (1934) did not consider that such a relationship existed. In *Tilletia holci* also, no such relationship could be observed. In some promycelia (Pl. I, fig. 9), as many as 15 nuclei could be counted before any sign of bifurcation of the promycelieum. But on the contrary, it appears that the development of the sporidia at the tip, or the bifurcation of the tip of the promycelium, starts after the completion of the haploid generation of nuclei in the promycelium.

Usually the primary sporidia are formed directly on the top of the promycelium (Pl. I, fig. 11, 12), but often the promycelium bifurcates, and each arm develops four sporidia (Pl. I, fig. 10). The number of primary sporidia is usually eight but it may vary from 4 to 16. The primary sporidia are long, acicular and slender, somewhat broad in the middle and tapering to a fine process at the end, but often blunt ended sporidia are also seen (Pl. I, fig. 14, 15, 16, Pl. II, 18). The mature primary sporidia measure 69—102×3—4μ. At the time of development of the primary sporidia, the promycelium puts out protuberances as small swellings at its tip. These continue to elongate giving rise to a cluster of primary sporidia; when young, they cluster together and their protoplasm is homogeneous. As the primary sporidia grow at the tip of the promycelium, the movement of the haploid nuclei towards the sporidia takes place in the promycelium. Often a crowd of haploid nuclei can be seen at the top of the promycelium below the young sporidia (Pl. I, fig. 11), or the haploid nuclei move more or less in a line one after the other (Pl. I, fig. 12). At some stage, the haploid nuclei enter the primary sporidia. Fully developed primary sporidia may still cluster together or they may fall apart. Usually the number of haploid nuclei corresponds to the number of primary sporidia, but often it has been observed that the number of nuclei is higher than that of the sporidia. Usually each sporidium received only one haploid nucleus (Pl. II, fig. 18), but binucleate sporidia are not uncommon (Pl. I, fig. 14). After the distribution of the haploid nuclei in the primary sporidia, any nuclei that may be left in the promycelium, remain there. When the nuclei have entered the sporidia and when they have attained a certain size, the primary sporidia are cut off by partition walls at their bases from the promycelium.

Soon after the entry of the nuclei, signs of conjugation appear between the primary sporidia. Communication between two sporidia is established by the formation of a short conjugating bridge at any point along the length of the sporidia but usually the connecting bridge is formed in the lower half. Sometimes two bridges are to be noticed between a fusing pair of sporidia. The nuclei of one sporidium passes to the other through this connecting bridge (Pl. I. fig. 16), giving rise to a binucleate primary sporidium. Often fusion between a binucleate sporidium and a uninucleate sporidium takes place giving rise to a trinucleate primary sporidium (Pl. I. fig. 14, 15). Conjugation between three primary uninucleate sporidia has been observed ; in such cases it appears that the nuclei and the protoplasm of the two pass to the third one. After the migration of the nucleus from one sporidium to the other, the cytoplasm from the non-nucleate sporidium flows to the binucleate one leaving the former empty. The empty sporidia are septate (Pl. I. fig. 16 Pl. II, 19, 23) like the empty promycelium. After the initiation of the binucleate condition in the primary sporidium, a bud begins to appear at some place in the binucleate sporidium (Pl. I. fig. 16), which soon grows and gives rise to a secondary sporidium. The secondary sporidia are attached to the primary ones by long or short stalks (Pl. II. fig. 19, 20, 21) from 3 to 15μ in length. Sometimes a swollen transparent structure, resembling a drop of liquid could be seen in the basal part of the secondary sporidium when the sporidia grow in liquid media, but in permanent preparations such swollen transparent structures are not seen. These structures may be drops of liquid similar to those found in the secondary sporidia of *Tilletia caries*, but in permanent preparations, probably in the process of killing, fixing and staining, the drops might have collapsed. As the secondary sporidia grow, the contents of the fused pair of primary sporidia migrate into it giving rise to a secondary binucleate sporidium (Pl. II. fig. 19). The secondary sporidia are distinguished by their shorter, wider, and somewhat curved

Text Fig. 4

and sickle-shaped appearance, resembling the secondary sporidia of *Tilletia caries*. Usually, the secondary sporidia are binucleate, but often they are uninucleate. These uninucleate secondary sporidia are developed from the unfused primary uninucleate sporidia (Pl. II fig. 20, 21), or sometimes from the binucleate primary sporidia. In *Tilletia caries*, Dastur (1921) has observed that most frequently the secondary sporidia contain a single nucleus, which he considered to be the result of the fusion of two paired nuclei. But on the other hand, Rawitscher (1914, 1922), Paravicini(1917) and Kharbush (1928) demonstrated that the secondary sporidia in *Tilletia caries* are binucleate. Buller & Vanterpool (1933), Wang (1934) and Holton & Heald (1941) hold that they may be uni- or binucleate. Here, in *Tilletia holci* nuclear fusion in the secondary sporidia was not observed. The secondary sporidia which develop from the primary binucleate sporidia are usually binucleate, but often a binucleate primary sporidium gives rise to a uninucleate secondary sporidium transmitting only one nucleus. Generally secondary uninucleate sporidia are developed from unfused primary uninucleate sporidia. Occasionally the secondary binucleate sporidia give rise to tertiary uninucleate sporidia (Pl. I. fig. 17). Often the binucleate primary sporidia instead of giving rise to secondary sporidia put out binucleate hyphæ (Pl. II fig. 22, 23). Sometimes, after the fusion, the contents of a primary binucleate sporidium come out as two secondary uninucleate sporidia (Pl. II. fig. 28).

Very soon the secondary sporidia become detached from the primary ones and begin to germinate, or they germinate while still attached to the primary sporidia. On germination they put forth hyphæ at one or both ends (Pl. II. fig. 21, 26, 27, 29, 30) or give rise to tertiary sporidia. The nuclear behaviour inside the secondary sporidia varies. On germination or before the germination both the nuclei may divide inside simultaneously or successively, giving rise to 3 or 4 nuclei (Pl. II fig. 25, 26, 29). On germination these nuclei migrate to the hyphæ, or both the nuclei migrate to the hypha where they divide successively or simultaneously (Pl. II fig. 31), or they continue to maintain the binucleate condition of the hypha for some time. Hyphæ that are produced from the secondary or tertiary sporidia are both bi- and multinucleate (Pl. II fig. 35). The nucleus of the uninucleate secondary sporidium also behaves in the same way ; either it divides inside the sporidium before, during, or after the germination, or it migrates to the hypha where it divides (Pl. II fig. 21).

The binucleate hyphæ which develop in place of the secondary binucleate sporidium, may grow for some time as a single strand or it may branch irregularly. The nuclei divide, supplying nuclei to these branches (Pl. II fig. 32). Hyphæ are nonseptate and branched, and contain two or more nuclei. Often these hyphæ give rise to uninucleate sporidia, which resemble the tertiary ones (Pl. II fig. 24).

In many preparations it has been observed that the nuclei of the secondary sporidia and the hyphæ are associated with one or two patches of chromatic vesicles, which are generally elongated (Pl. II fig. 33, 34).

Study of the Fungus in the Host Tissues

Holcus mollis is a perennial grass. The mycelium of *Tilletia holci* perennates in the rhizome. Longitudinal and transverse sections of the rhizome show that the intercellular spaces of the parenchymatous cells, and sometimes the tissues of the vascular elements, are infected with long, branched and imperfectedly septate, vegetative hyphæ. Along with the development of the stem-buds from the rhizome the hyphæ migrate to the young buds, and keep pace with the growth of the haulms.

The mycelium moves about in the intercellular spaces of the ground tissue of the stem, and often along the elements of the vascular bundles and finally come to the panicle. In the sections of a young ovary, one finds that the vegetative hyphæ are dispersed here and there in the intercellular spaces of the paranchymatous cells. These hyphæ are much branched, long, and ramify in all directions (Text fig. 3). Hyphal cells are fairly long (8—12μ long and 1—2μ broad) binucleate or pluri-nucleate. The protoplasm of the fungus is always much more densely stained than that of the host. The nuclei of the parasite are small, and appear as small chromatic corpuscles surrounded by a small clear zone. The protoplasm is more or less hollowed by the presence of vacuoles of various size and shape.

In the young ovary the mycelium is very abundant, especially in the central zone, ramifying in all directions in the intercellular spaces and occasionally inside the cells. The hyphæ are thin, and contain two or more nuclei in each hyphal cell.

DEVELOPMENT OF THE CHLAMYDOSPORES IN THE HOST PLANT

At the time of spore formation, the ends of the hyphæ enlarge a little and these enlargements usually contain two nuclei (Pl. II fig. 36, 37, 38). The cellular contents gradually accumulate in these swellings which are cut off by walls from the rest of the hyphæ. As these swellings enlarge they give a blister like appearance. In some of the swellings one can see only one nucleus, but the size of the nucleus suggests that this might be a fusion nucleus. Usually the nuclear fusion occurs when the spores are quite young, but in certain cases the fusion is delayed for some time. As the young spores gradually and progressively enlarge their nuclei approach one another and finally fuse (Pl. II fig. 39, 40, 41). The fusion nucleus enlarges along with the increase of size of the spore. The young spore has protoplasm, dense and homogeneous in appearance. Later on vacuoles appear, and in a mature chlamydospore one can easily distinguish one to several fairly large vacuoles. At a certain age the protoplasm of the spore secretes small droplets of oil which are distributed irregularly and are coloured black by the stain. Up to a certain size—about $\frac{3}{4}$ the diameter of the mature spore—the nucleus and the cell contents remain visible in the stained preparations, after which irregular thickening, in the form of reticulations appear on the wall (Pl. II fig. 42). In the mature spore the reticulations become very prominent, and the colour of the outer wall gradually changes from transparent to olive brown and then to dark brown. The depth of the reticulations varies from spore to spore as already noted.

The development of the spores follows a centrifugal direction. While the spores mature at the central region of the ovary, they are surrounded by spores proportionately younger, and still further away, that is near the ovary wall, hyphæ could be seen still cutting off young spores at their tips. Finally the whole of the ovarian cavity is filled up with spores.

PATHOLOGICAL MODIFICATIONS OF THE INFECTED HOST TISSUE

The nucleus of a healthy cell of *Holcus mollis* is quite round, oval or elongated in contour, and contains one to more deeply staining nucleoli. The nucleoplasm is stained fairly deep grey by hæmatoxylin. The infected cells of the rhizome, stem and inflorescence axis are more or less compatible with the parasite ; that is, the host cells seem to maintain their healthy condition though the hyphæ are distributed in the intercellular spaces. However, certain cells seem to be hypersensitive, and,

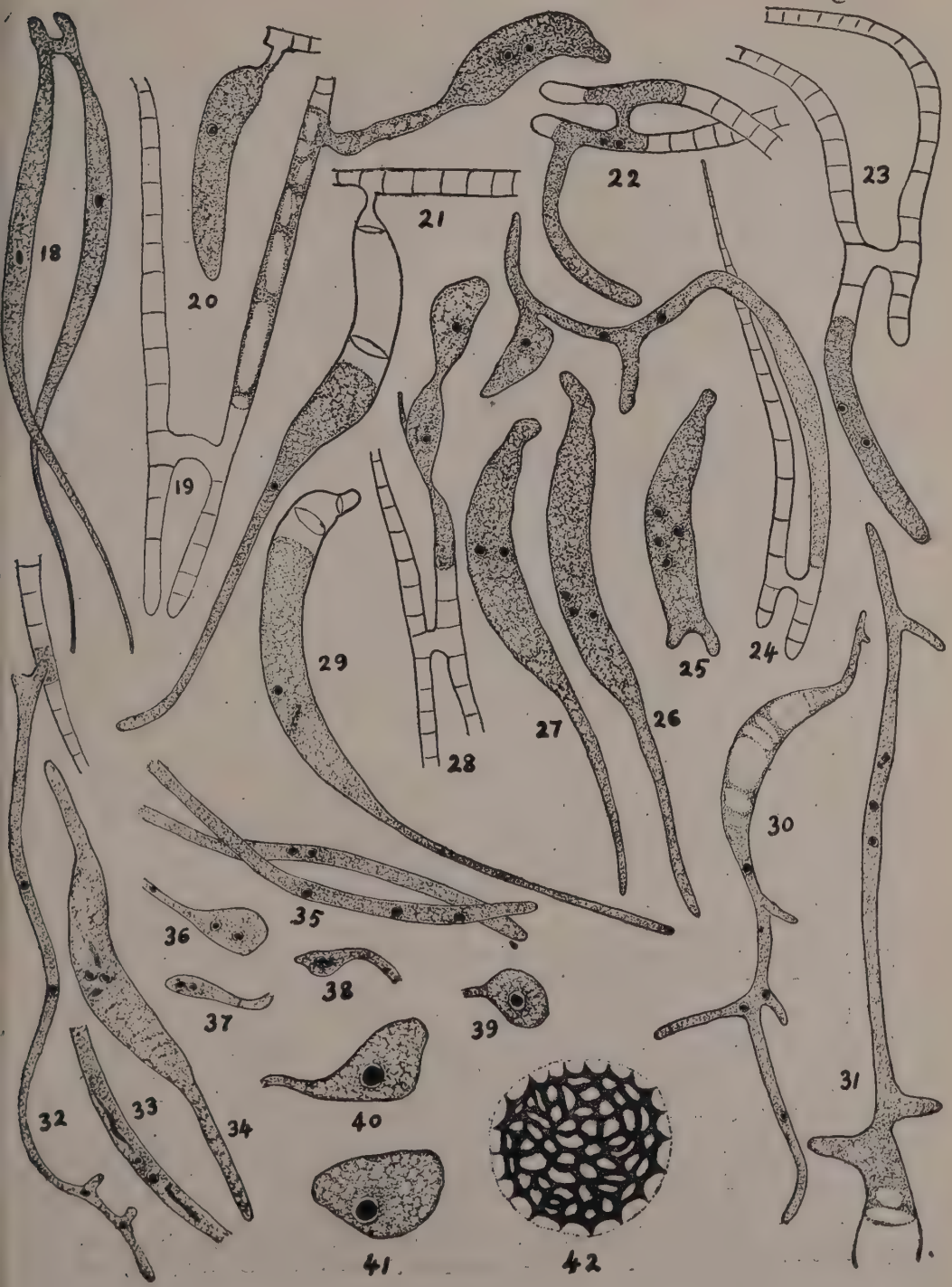


Plate II. Figs. 18 to 42. See legend for explanation

before the invasion by the parasite, the nuclei begin to degenerate. Most frequently the nucleoplasm becomes more and more deeply stained, until one can see only a black mass without any distinction of nuclear membrane or nucleoli. This black mass gradually loses its contour and disintegrates into irrgeular masses dispersed in the cytoplasm of the cell. The cell cytoplasm also often becomes deeply granular, and finally turns into a black mass which soon falls into bits and is ingested by the healthy cells or by the parasite. Sometimes the degeneration of the nucleus is revealed by the disappearance of the nuclear membrane and the nucleoli.

Marked degeneration of the cells in the infected ovary is noticed when the hyphæ are in the intercellular spaces or inside the cells. In the normal state, the cells of the ovary contain numerous leucoplasts which are more or less elliptical and have black granules inside. In the infected cells they become pale, and gradually decrease in size and finally disappear. The nucleus gradually loses its contour and turns black (Text fig. 4). The cellular membrane becomes perforated and soon breaks down. Amongst the mycelium, in the process of spore formation, one can find the traces of the remains of cellular constituents of various size and shapes. Ultimately these are ingested by the hyphæ, and the whole ovarian cavity is replaced by the chlamydospores.

DISCUSSION

Cytologically *Tilletia holci* bears a very strong resemblance to *Tilletia caries* and *Tilletia fœtida*. In *Tilletia holci* the chlamydospore on germination gives rise to a promycelium which develops a whorl of sporidia at its tip. The growth of the promycelium varies according to the nature of the division of the diploid nucleus. After completion of all the nuclear divisions the sporidia are developed. The usual number is eight, but it varies from four to sixteen.

Since the discovery of Dangeard (1892), it has been established that the chlamydospores of species of *Tilletia* are diploid in nature. According to the individual peculiarity, the behaviour of the diploid nucleus varies at the time of germination. Holton and Heald (1941) reviewing and discussing the work done on *Tilletia caries* by various investigators, shared Wang's (1934) view, that the diploid nucleus of the chlamydospores may complete all its divisions either inside the spore or in the promycelium with all the possible intermediate stages. In *Tilletia holci* the nature of divisions of the diploid nucleus is similar to that of *Tilletia caries* as observed by Wang (1934). The diploid nucleus may complete all the divisions inside the spore or in the promycelium or in both.

The promycelium, while the nuclear divisions are taking place inside it, keeps on elongating until all the nuclear divisions are completed within it. Only then do the primary sporidia develop at the tip of the promycelium. On the other hand, when the nuclear divisions are completed inside the spore, the promycelium after a short period of growth develops sporidia.

It is determined for the first time in *Tilletia holci* that the diploid number of chromosomes is four and the haploid number is two.

According to Paravicini (1917), the number of sporidia in *Tillitia caries* corresponds to the number of haploid nuclei in the promycelium. But Dastur (1921) and

Wang (1934) demonstrated that the numbers do not always correspond. In *Tilletia holci* the number of sporidia is usually equal to the number of haploid nuclei, but often the number of nuclei is more than that of the sporidia, and rarely the number of sporidia is greater than the number of nuclei.

As observed by Rawitscher (1914) in *Tilletia caries*, the primary sporidia of *Tilletia holci* also fuse in pairs, and the nucleus from one sporidium migrates to the other, giving rise to a primary binucleate sporidium. According to Hanna (1934), the secondary conidia of *Tilletia caries* and *Tilletia fœtida* are uninucleate but in *Tilletia holci* the secondary sporidia are usually binucleate and always developed from primary binucleate sporidia. Only occasionally do the primary binucleate sporidia produce secondary uninculate sporidia transmitting only one nucleus. The secondary uninucleate sporidia may also be developed from unfused primary uninucleate sporidia. These observations agree with those of Buller and Vanterpool (1933), Wang (1934) and Holton and Heald (1941), made in *Tilletia caries* and *Tilletia fœtida*. Dastur (1921) claimed to have observed a few cases of "the fusion of the conjugate nuclei" in the secondary sporidia of *Tilletia cariss*. He further stated that "the presence of the two nuclei in the secondary sporidium is, at least in some cases, due to the division of a single non-conjugate nucleus." In the present investigation, no fusion of the conjugate nuclei has been observed in the secondary sporidium. The secondary sporidia are always binucleate or uninucleate depending on their origin. Furthermore, on the germination of the secondary sporidium, the pair of conjugate nuclei migrate directly to the hyphæ where they divide, or sometimes both the nuclei divide successively or simultaneously inside the secondary sporidium, at the time of germination of the latter, than migrating to the hypha. The single nucleus of the secondary uninucleate sporidium also behaves in the same way. Often however, the primary binucleate sporidium gives rise to dicaryophytic hypha directly, instead of producing secondary sporidium.

The parasitic mycelium is mostly intercellular, and occasionally intra-cellular. The vegetative mycelium is irregularly branched and septate. Hyphæ move along the inter-cellular spaces of the host tissues in all directions. The nuclear condition of the parasitic mycelium is similar to that of *Tilletia caries* as observed by Kharbush (1928). Most of the hyphal cells are binucleate but occasionally pluri-nucleate cells can also be observed.

Chlamydospores are exclusively formed in the cavities of the ovaries of the spikelets. The developmental history is similar to that of *Tilletia caries* as described by Wang (1934) and Kharbush (1928). Tips of the sporiferous hyphæ which are binucleate, swell and enlarge. Nuclear fusion takes place, giving rise to young spores. A mature spore always contains a large sexual nucleus and has an outer wall which is deeply reticulate.

In stems and rhizomes, cells in the presence of mycelium seem to preserve their constituents in the normal state. In the ovaries signs of degeneration appear after some time. The first sign of degeneration appears in the nucleus, which gradually loses its deep colour and decreases in size. The nucleoli disappear, and finally they break up into masses of irregular chromatic accumulations. The cytoplasm often disappears gradually. Sometimes it at first contracts and then breaks up into small masses. These elements gradually become reduced and disappear amongst the mycelium of the parasite.

SUMMARY

1. *Tilletia holci* forms brownish-black loose spore masses in the ovaries of *Holcus mollis*. Spores are yellowish brown to deep brown with deep reticulations on the epispores.

2. Mature chlamydospore contains a diploid nucleus.

3. Under ordinary conditions, the germination of the chlamydospore is delayed. Spores bleached with 10% bleaching powder solution and kept outside in sterile water in hanging drops and in 1 per cent agar at temp.—1°C to 3°C for about 5 to 7 days, and then removed to the room temp. of about 18°C yielded about 50 to 80 per cent germination within 48 hours.

4. On germination the chlamydospore puts out a long or short promycelium, at the tip of which a cluster of sporidia are developed. The number of sporidia varies from 4 to 16.

5. The diploid nucleus of the chlamydospore may complete all the divisions inside the spore or in the promycelium or some in the spore and some in the promycelium, resulting in the production of final generation of haploid nuclei.

6. In the promycelium, the first division of the diploid nucleus is equational and the second one is reductional.

7. Diploid number of chromosomes is 4 and the haploid number is 2.

8. Usually the haploid number of nuclei corresponds to the number of primary sporidia, but often the number of nuclei is greater than that of the sporidia.

9. Usually each primary sporidium receives only one haploid nucleus.

10. Fusion between the primary sporidia is accomplished by a short conjugating bridge. The nucleus of one of the fusing pair of sporidia passes to the other giving rise to a primary binucleate sporidium.

11. Usually the primary binucleate sporidium gives rise to secondary binucleate sporidium, but sometimes it produces a uninucleate secondary sporidium transmitting only one nucleus.

12. Secondary uni- or bi-nucleate sporidia germinate directly giving rise to hypha.

13. Parasitic mycelium in the tissue of the host plant is irregularly branched and septate. Hyphal cells are binucleate to plur-inucleate.

14. At the time of chlamydospore formation, the end cells of the binucleate sporiferous hyphæ swell, forming blister like structures. Nuclear fusion takes place in them and ultimately they are transformed into chlamydospores.

15. Chlamydospores are exclusively formed in the ovaries.

16. Infected cells, especially in the ovaries of the spikelets where the fungus fructifies, are degenerated and finally disappear.

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EXPLANATIONS OF PLATES

(All figures were drawn with camera lucida at X 3000, except figure 10 which was at 2100.)

PLATE I

Fig. 1. A chlamydospore showing the diploid nucleus, vacuoles and the dark staining bodies inside the spore.

2 & 3. Germinating spores showing several nuclei inside them before the development of promycelia.

4. Germinating spore showing the protrusion of promycelium and migration of nuclei.

5. A binucleate stage in the promycelium. Note that the upper nucleus is in a state of anaphasic separation. 4 chromosomes are divided into 2 groups, each group containing 2 chromosomes.

6. A germinating chlamydospore showing 8 nuclei in the promycelium.

7. The diploid nucleus has migrated into the promycelium and there it has resolved to 4 chromosomes in preparation for the first division.

8. Four-nucleate stage in the promycelium.

9. A long promycelium showing 15 nuclei inside it.

10. Bifurcation of the promycelium. Note that each branch has developed 4 sporidia.

11. & 12. Development of sporidia at the tips of the promycelia and the movement of the nuclei inside the promycelia towards the sporidia.

13. Branched promycelium, showing the development of sporidia from one of the branches.

14. Conjugating sporidia, showing two nuclei in one sporidium and one nucleus in the other sporidium.

15. A stage after the migration of the nucleus from the uninucleate sporidium to the binucleate one giving rise to a trinucleate sporidium.

16. Conjugating sporidia, showing the migration of the nucleus and the cell contents from one sporidium to the other resulting in the development of a primary binucleate sporidium. Note also that the binucleate sporidium is about to develop a secondary sporidium.

17. Development of uninucleate tertiary sporidium from a binucleate secondary sporidium.

PLATE II

18. Two uninculate primary sporidia are in a state of conjugation.

19. Development of secondary binucleate sporidium from primary binucleate sporidium.

20. Development of secondary uninucleate sporidium from a primary uninucleate sporidium.

21. Germination of secondary uninucleate sporidium.

22 & 23. Direct development of hyphæ from binucleate secondary sporidia.

24. Development of a hypha and from there sporidium from the binucleate primary sporidium.

25—27. 4—, 3— and 2—nucleate stage of the secondary sporidia at the time of germination.

28. Development of two uninucleate secondary sporidia from a primary binucleate sporidium.

29. Germination of a secondary binucleate sporidium. Note that one nucleus is in a state of division.

30. Developments of hyphæ from both ends of the secondary sporidium, but the hypha at one end continues to grow which later on branches.

31. A hypha developed from the secondary sporidium. Note that one nucleus inside it is in a state of division.

32 & 35. Hyphæ showing the bi-and pluri-nucleate conditions.

33 & 34. A portion of a hypha and a secondary sporidium showing the chromatin vesicles associated with the nuclei.

36 & 41. Various stages of the development of chlamydospores.

42. Development of reticulations on the epispore.

Text figure 1. x 4

Portion of a smutted panicle of *Holcus mollis*. Ovaries are converted to sori filled up with deep brown to olive brown spores.

Text figure 2. x 650

Chlamydospores of *T. holci*. The upper three spores are deep brown with low reticulations and bigger inner diameter. The lower three spores are olive brown and have comparatively smaller inner diameter and high reticulations.

Text figure 3. x 3000

Cells from the young ovary of *H. mollis* infected with *T. holci*, showing the hyphæ in the intercellular spaces.

Text figure 4. x 3000

Cells from the infected ovary of *H. mollis* parasitised by *T. holci*, showing the degeneration of the host cells.

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